Filtration and reuse of water in fish farming





PREFACE

This book has been developed by biologists at Hydrotech AB in Sweden, a leading company in filtration techniques. We would like to inform our customers how our filters can be adapted as an integrated part of re-use systems, and as end of line facility treatment to improve water quality.



Hydrotech AB was founded in 1984, and has ever since been a market leader in microscreening of water in aquaculture facilities. The first filter developed for filtration with microscreens was the Triangelfilter, which was sold in more than 1500 units. Since then Hydrotech developed Drumfilters and recently a new invention, the Discfilter. This wide range of filters covers the specific needs of each fish farmer. By writing this book we hope that people interested in fish farming can learn something about re-use of water and maybe in this way avoid too many mistakes, as our clients success is important for us and for the business in general. However, Hydrotech AB is not responsible for sizing of systems based on information in this book. Information must be considered as guidelines. Text or pictures must not be copied without prior written notice



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1. DEFINITION OF RECIRCULATION

A traditional fish farm with ponds or tanks, has an inlet and outlet, and water is used a few times or it's a simple flow through system. If water is treated to improve quality and used again, then it's considered a re-use or recirculation system.

However, as simple it seems to define a recirculated aquaculture system (RAS), the more complicated it seems to have common nominations as how to evaluate the technology behind.

The purpose of this booklet is to describe water purification technologies used in aquaculture facilities, with special emphasis on microfiltration.



The definition below is very often seen:

DEGREE OF RECIRCULATION: (1 - A ÷ (A+C)) X 100 = % RECIRCULATION

In practical terms if e.g. 10 l/s is added to the system as fresh water and 90 l/s is recirculated, the resulting degree of recirculation will be 90 %.

A wide range of equipment can be implemented to increase recirculation of water, as can be seen from the figure below:



Figure 2. Design of system and re-use of water

A high degree of recirculation is not an adequate measure on the capacity of any RAS, e.g. if a large volume of water is recirculated with a very low stocking density, it can be recirculated without any cleaning, so it appears very efficient with a traditional definition of recirculation.

A typical closed system will have a degree of recirculation of more than 99 %.

A better design and cost related factor is recommended as a supplement for evaluation of closed systems:

m³ fresh water per kg of feed added to the system

This factor will reveal more about the technical level of the system. A typical range of closed systems are from 20-500 l of fresh water per kg of feed applied to the system.



Picture 1. Feeding eels

2. COMPONENTS OF RECIRCULATED SYSTEMS.

Tanks or ponds:

The aquatic environment must meet the needs of the farmed organism, both in respect of water quality and physical properties.

Earth ponds are very widespread, mainly because of cost effective construction.



Picture 2. Traditional Danish trout farm

Farmed for more than 100 years, rainbow trout is well adapted to intensive culture. In the beginning of this century, feeding of trout was infrequent and natural food sources were a large part of their diet. Early trout culture relied on low stocking densities. Where natural conditions with soft ground prevailed, the pond system was preserved. Today, advances in nutrition and equipment technologies permits trout farmers to increase stocking densities and efficiencies.

In the last 20 years a much more "industrialised" form of fishfarming has been developed; especially salmon farming in Norway, eel farming in Holland, Denmark, Taiwan and other S.E. Asian countries. Leaving trout and catfish aside, tilapia and stripped bass are the predominate species cultured in the USA, where key factors are:

- High value species
- Fresh fish to the market
- high output per m²
- production independent of water source



Picture 3. Recirculated smolt farm in Denmark

Reuse of water is an inevitable consequence if one is to meet the demands above. Design of rearing units must be considered to meet the needs of the fish. Selfcleaning properties, cost and space considerations are just a few of the criteria to determine optimum choice. There are some basic constructions:



Figure 3. basic tank constructions

There are other types of construction, but above are the most practical and widespread constructions.



Picture 4. Trout farm in Spain.

Choice of tank must reflect:

- Fish species, is it bottom dwelling fish ? (as i.e. eels and salmon smolt) or rainbow trout, where the whole water column will be used? Square meters (m²) are used for the first type(and as economic evaluation) and cubic (m³) are used for sizing of the latter type.
- Space available?
- Self-cleaning properties?
- Regulation of water current and oxygen transport ?

Properties	Round tank	Square, round corners	"Race-ways"
self cleaning effect	5	4	3
residence time of particles	5	4	3
regulation of oxygen	5	5	3
use of space	2	4	5

For evaluation of tank properties, see table below:

Ratings from (1-5), 5 is the best feature and 1 the lowest.

As can be seen from above, there is no easy answer for selection of tanks based on physical properties; biology of fish species must also be considered. Self-cleaning tanks will improve water quality in the tank, and increase efficiency of down-stream filtration. Feces and food pellets will leak organic material (BOD), nitrogen and phosphorous within a few minutes. In a round tank, particles have a residence time of 2-3 minutes, due to the hydraulic pattern and gravitational forces.



Figure 4. Vertical inlet with horizontal adjustment

A vertical inlet with horizontal adjustment, is very efficient for control of current in the tank, but must not exceed app. 0,3 m/s. This is accomplished by making several holes in the vertical portion of the pipe.

Note: If pumps are placed lower than water levels in tanks, remember to drill a small hole above water surface, otherwise water might run by gravitation from tank to reservoir, emptying the tank.

In raceways there is no impulse force to create a current, velocity in the tank is simply calculated from water flow in a channel:

$$\mathbf{Q} = \mathbf{A} \times \mathbf{V}$$

(Q = water flow, m^3/h . A = cross section area, m^2 . V = velocity of water, m/s)

Only by pumping more water to the tank, can the current be controlled. To obtain self-cleaning properties a velocity at the bottom of 4-5 cm/s will be preferred to transport particles. For all types of tanks, stocking density of fish is very important, as the fish will have a "cleaning" effect by their activity. For all types of tanks, the inclination of the bottom has less importance, 2-5 cm/m are normally recommended, but this is mainly for proper draining of the tank.



Picture 5. Round tanks in an eel farm in Holland.

Some fish species are rheophile (they prefer moving water). Tests prove that streaming water improves their growth and feed conversion. Some salmonides are actually "body-building" when they are swimming at a certain speed, approx. one body length per. Sec. (depending on size!), while other species such as the eels are typically "bottom-dwelling" fish. Outlets of tanks must be constructed for optimum transport of particles. Water velocity must not be too high (approx. 0,3 m/s), and simultaneously hole sizes of tank drains must be small enough to prevent fish escapement.



Figure 5. Gravitational outlet:



Picture 6. Self-cleaning gravitational outlet.

This model is mainly used for eels. This design facilitates ease of tank draining. Dead fish will end up on a grid at the surface. This assembly is normally mounted with a motor and brush, turning slowly. Holes for small eels are down to a length of 0,5-1,0 mm. The outlet must be self-cleaning. With larger holes the self-cleaning device may not be necessary and the grid structure could be placed horizontally.

3. WASTE FROM FISH FARMING

All waste originates from the feed added to the system. Modern dry feed is manufactured for individual species, but in general all feed today is high energy.



Figure 6.



For proper sizing of down stream water treatment and water conditioning, you need to know what type of waste is produced and in which amounts?

One kg of dry feed for i.e. salmon or eel is approximately composed as follows:

1 kg feed	Content	Energy content (g).	Total energy content
Protein:	500 g	5,65	2825
Fat	250 g	8,30 *	2075
Carbohydrates:	120g	4,1	492
Total gross energy			5392

* 9,45 kcal/g as can be seen on most fish feed labels are too high, due to high content of fish oil (unsaturated fatty acids) it will be lower.

Nitrogen is contained in the proteins, approx. 16 % based on weight. The organic loadings are derived from protein, fat and carbohydrates and are normally expressed in indirect terms as chemical analysis is difficult. It can be estimated as a biochemical oxygen demand (BOD) or by chemical oxygen demand (COD).

For production of 1 kg of fish, the waste loading can be calculated at different feed conversion ratios, if it is assumed that feed type and content of protein, fat and phosphorus are constant in fish and feed.

The resulting loading can be expressed as a linear relation to the feed conversion:

Max loading, tot. N(g. N per kg feed) = 80 x feed conversion - 30

Equation 1.

Nitrogen: Content in feed as above, small trout will have a content of protein at approx. 19 %. This is not a physiological term. It can only be used within a normal range of feed conversion ratios. However, it can be used as a guideline for loading of nitrogen and thereby sizing of biofilters.

Organic loading:

Respiration i.e. consumption of oxygen, yields energy to the organism. Organic material is degraded and energy transferred to ATP, which is the current form of energy in the cells. Oxidation of each gram of protein, fat and carbohydrates yields different amounts of energy, but as an average, on a carnivorous diet, it has been calculated that a conversion figure of 3,24 calories correspond to 1 mg of oxygen.

COD: Chemical Oxygen Demand, (based on above) can be calculated as follows: 1 kg feed: $5.392 \text{ kcal} / 3,24 = 1.664 \text{ g O}_2$

The calculation of final potential outlet of organics from tanks, is not so easy because some of the food is used for respiration. Based on the energy equation, fate of energy can be described as:

$\mathbf{C} = \mathbf{G} + \mathbf{R} + \mathbf{U} + \mathbf{F}$	
C = consumption, G = growth, R = respiration, U = Excretion, F = feces	

This energy equation is based on physiological measurable terms, and as can be seen from the calculation below, loss of feed and thereby calculation of actual feed intake is very difficult to measure accurately.

COD, in 1 kg of fish (small trout), 19 % protein, and 8 % fat.

1 kg fish	Content	Energy content (g)	Total energy
Protein	190 g	5,65	1074
Fat	80 g	8,30	664
Total gross engergy, kcal			1738

Fish are very efficient converters of energy approx. 30 % of energy intake, will be transformed into body growth. The difference between growth and feed intake is energy consumed for respiration, which is intimately connected to the costs of growth. Respiration is estimated at 70 % of potential oxygen loading.

Fate of feed expressed as COD from 1 kg feed:

Feed::	1.664 g O ₂
Respiration::	790 g O ₂
Growth::	536 g O ₂
Feces and feed loss::	338 g O ₂ ²

(figures can be changed according to fish species and feed type)

Waste feed and feces makes up the organic loading on microscreen filtration and biological filters. A quantitative term for organic material often used is Biochemical oxygen demand (BOD), but it's not an exact measurement, as the oxygen consumption describes the intensity of a microbiological process. In some tests it has been shown that about 50-70 % of fish feed and waste is easily bio-degradeable and thereby measured as BOD. The non-degradeable organic material will to some extent accumulate in the system, resulting in a brown "tea like" color.

Based on above assumptions, following potential loading can be expected per kg feed used in the farm:

Potential loading of 1 kg feed in the system: 169-237 g BOD

In the table below, efficiency of a microscreen in terms of mass balances in a RAS, has been calculated. :

Parameter	30 micron filter	60 micron filter	100 micron filter
Suspended solids	75 %	36 %	16 %
Total nitrogen	62 %	29 %	14 %
Total phosfor	43 %	39 %	20 %

Removal efficiency of different mesh sizes of a Hydrotech microscreen as a percentage of particulate matter directly originating from feeding. "Blue label" project, Fair-CT-98-9158.

Above data only relates to eel farms, figures may be different on various species! It can be seen that 30 microns is significantly better than coarser screens.

4. PARTICLE FILTRATION

Particles are the undissolved fraction of the feed input to the system. These particles are normally measured as suspended solids (SS). In a recirculated fish farm, particles originate from waste feed and feces. In a farm with intake from surface waters, organic and inorganic particles will also constitute a portion of the particle distribution.

As stated in the section on tanks and ponds, self-cleaning properties are very important for down stream filtration systems, particles must be transported quickly and removed gently from the system.

Component	Circular tanks	Raceways	Pond systems
Tot. P	70-80 %	50-70 %	30-50 %
BOD	70-80 %	60-70 %	40-60 %
S.S.	80-90 %	60-80 %	40-60 %
Tot. N	20-50 %	20-40 %	20-40 %
NH_3/NH_4^+	-	-	-

See table below for average removal efficiencies in different types of fish farms:

Above efficiencies are estimates of Hydrotech Drumfilters applied with 60 μ m filtercloth.

Microscreens can be installed at fish farm outlets for reduction of solids loading on downstream rivers.Sometimes filters are employed at inlets to increase intake water quality. Lickey Bridge smolt farm in Ireland. Occasional flooding caused problems when the water contained a lot of clay and sand:

Diameter range (µm)	Mg / L
Total	114
> 63	34
38-63	14
20-38	16
11-20	23
5,5-11	17
2,2-5,5	10
< 2	0.07

Particle size distribution, inlet water Lickey Bridge.

Based on this data, a 20 μ m filter cloth was chosen. This is very fine filtration for a filter based on gravitation.



Picture 9 Disc filter, HSF2106 at Lickey Bridge:



Picture 10. Recirculated fish farm in Denmark. Water is gravitated from tanks to microfilter.



Picture 11. Drumfilter, model HDF1606-1B

Filtration in recirculated systems:

Hydrotech Drum- and Discfilters are in widespread use all over the world in recirculated systems. Their unique design of filtercloth, secures a gentle removal of particles: (See added leaflets on filters).



Function of Drum- and Discfilters:

- 1. Water to be filtered enters the inside of the drum.
- 2. The water is filtered through the drum's filter elements. The difference in water level inside/outside the drum is the driving force for the sieving.
- 3. Solids are trapped on the filter elements and lifted to the back-wash area by the rotation of the drum. The drum rotation is intermittent or continuous depending on type of control.
- 4. Water from rinse nozzles is sprayed from the outside of the filter elements. The rejected material is washed out of the filter elements into the sludge tray.
- 5. Rejected sludge flows together with water by gravity out of the filter.

In a recirculated system, microfiltration is an integral part of a complete purification system.

The benefits of microscreen filtration are as follows:

- Reduction of organic loading on biofilter(s).
- Improving turbidity of water.
- Reduction of larvae and eggs of some parasites.
- Improving conditions for nitrification
- Stabilising function of biofilters.
- No "wild" spreading of larvae, i.e. Tilapia.

The most cost effective way to get rid of waste is by mechanical filtration i.e. low energy and no oxygen consumption for elimination of impurities. The normal range of filtration is $20 - 100 \mu m$.

As density of suspended solids (SS) in a recirculated system is very low, approx. 1,1-1,2 g/cm³, settling is not feasible. Filtration has proved to be the only practical solution.

Approx. 50 % of BOD will be removed in a proper sized filtration system. The remaining part of BOD will be dissolved and must be absorbed and digested in the biofilters.

The subsequent loading of biofilters by organic material will result in growth of bacteria. Abrasion of the formed "bio-film" will decrease turbidity of the water. The formed particles are very small. See the figure below on relative particle distribution in a system with and without a microscreen:



Figure 9. Relative distribution of particles

If microscreens are included in the system, the majority of the solids in the rearing units will be removed. Suspended solids are present as small parts of dead bacteria, less than $20 \ \mu m$.

Water consumption:

The sludge from micro filters has to be removed from the system and consumption of water for the rinse water system, should be as low as possible

Filter opening	30 micron	60 micron	100 micron
Litre/kg feed	200	100	50
% of recycled water	0,23	0,11	0,06

Relative water consumption for a drumfilter at 30, 60 and 100 micron filter opening

As could be expected the relative water consumption as well as the fraction of the recycled water used for spraying is decreasing with increasing mesh size from 200 to 50 litre per kg feed and 0,23 to 0,06 % of the internal flow. This is acceptable levels of water exchange in a RAS system, if needed the water consumption may be further reduced by including sludge concentration in the system.

5. BIOLOGICAL FILTRATION

Some of the waste from fish and feed will be dissolved and has to be degraded or transformed into harmless substances. Again you have to consider organic material and nitrogen containing substances, (Phosphorous is an inert substance, with no toxic effect).

Sizing of biological filters must be based on expected loading of organic material and nitrogen compounds.

Organic material.

The term BOD, includes most of the bio-degradable material as proteins, fat and carbohydrates, at degradation by Heterotrophe bacteria in an oxygen rich environment, below simplified reaction will take place:

 $\begin{array}{ccc} \mathrm{C_6H_{12}O_6} + \ \mathrm{6O_2} & \Rightarrow & \mathrm{6\ CO_2} + \ \mathrm{6\ H_2O} + \ \mathrm{E.} \\ (\ \mathrm{glucose} \) & & (\ \mathrm{carbondioxide} \) & (\ \mathrm{Energy} \) \end{array}$

The bacteria will grow through multiple divisions, this is called the "yield", which is quite high approx. 0.6 kg / kg BOD. This "bio-film" or rather sludge, will end up as small particles (as described above) and at proper management it can be backwashed from bio-towers.

Bacteriological reactions are very temperature and pH sensitive, in a recirculated eel farm at 25 °C, degradation of organics in biofilters, will be: <u>Approx. 10-15 g</u> <u>BOD / $m^2 \times day$ </u>.

Organic material may be degraded without available oxygen present--fermentation.

In sizing of biofilters these reactions have normally not been emphasised, but if mismanaged it may cause problems and water quality deterioration. There have been problems in sub-merged filters, even if oxygen is present at outlet, short circuits may appear due to accumulation of sludge, and thereby formation of pockets without oxygen.

Fermentation process:

Organic material \Rightarrow H₂, CO₂, alcohol, acetic acid \Rightarrow Methane(CH₄) and other org. acids

It's a multi-step reaction and very sensitive, especially the second step i.e. a small change in pH may severely reduce transformation of alcohols, organic acids etc., which will cause accumulation of easy degradable material in bio-towers. A sudden change in physical properties may release accumulated organics, and as it comes in contact with oxygen in reservoirs and fish tanks, it will, in a very short time be degraded by heterotrophe oxygen consuming bacteria---sometimes so fast that oxygen supply can't sustain oxygen consumption. It's virtually an "organic bomb". This situation has to be avoided. Proper physical sizing and management of filters do this, which prevents this accumulation.



Figure 10. The nitrogen cycle

These nitrifying bacteria are called autotrophe bacteria because they have an inorganic compound, ammonia, as an energy source. The process has a much lower yield (growth of bacteria) than the heterotrophe bacteria, app. 10 %. The process needs oxygen (aerobic) and produces H^+ (acid), lowering pH.

As can be seen from figures below, reaction kinetics are very dependent on environmental factors:

A: Temperature.

Temperature ° C	5	10	15	20	25
Nitrification, (g NH_4 -N / $m^2 \times d$)	0,3	0,5	0,7	0,9	1,0

Figures are at optimum conditions at given temperature.

B. Organic loading.



Figure 11. Organic loading

Competition on available space in biofilters are not in favour of nitrifying bacteria, due to the slow growth, compared to heterotrophe bacteria.

C. Oxygen concentration.



The transfer of oxygen to bacteria must be controlled. Outlet water of submerged filters, must have a concentration of $3-4 \text{ mg O}_2/\text{L}$ as minimum .

It's recommended to run the filters as "nitrification" filters i.e. secure optimum conditions for nitrification. This should take into consideration the above factors: low loading of organic material and ample oxygen supply. pH is also very important, but the process seems to be quite stable within limits of pH 6,5- 8,0. For all environmental factors, a sudden change is much worse than a slow change as bacteria can adapt within a certain range.

Calculation of surface area in biofilters:

Above mentioned processes must be considered:

- Biodegradation of organic material.
- Nitrification.

Organic loading has been calculated to be max 237 g BOD per kg feed applied to the system. Efficiency of microfiltration can be estimated to 50 % reduction of organic particles. Resulting sizing figure on BOD loading on biological filters, is therefore max 119 g per kg feed. From equation (1), at a feed conversion of 1, it can be seen that the corresponding production of Tot. Nitrogen will be 50 g -N.

BOD: 5 g / m² x day, 119 g BOD / kg feed-----: 24 m² Ammonia: 0,7 g / m² x day, 50 g NH₃/NH₄-N----: 71 m²

At this low loading of organics, there will be enough space available for nitrification: Total specific surface area in biofilters----- = 95 m^2 (for each kg feed / day, used in the system)

Denitrification.

Due to nitrification, nitrate will accumulate in the system. Levels of nitrate must not exceed approx. 90 mg NO_3 -N / L. Such high levels seems to have an impact on growth and feed conversion. It has been considered quite harmless, but it seems that some fish species are affected by too high concentrations.

The process of reducing nitrate to atmospheric nitrogen is called denitrification. This process requires an organic source and will take place in anoxic (without oxygen) environments. The predominate denitrifying bacteria is pseudomonas.

5 CH₃OH + 6 NO₃ \Rightarrow 5 CO₂ + 3 N₂ + 6 OH + 7 H₂O (methanol)

In practical terms you need approx. 2,5 kg CH₃OH for each kg NO₃-N. Sludge from microscreens has been used as organic source, but it seems that excessive accumulation of sludge takes place, resulting in back-washing problems. In submerged filters extensive denitrification will take place, even though the filters are operated as aerobe filters, this is due to the fact that the "bio-film" will be in a state of anoxic conditions in the deeper layers. But it can't be recommended to try to monitor all processes on-line i.e. oxidation of organic material, nitrification and denitrification. Denitrification must be operated in a "by-pass" to avoid accumulation of organic material in filters designed for nitrification. Typical design of a denitrification tank: A tank with filter media (100 m²/m³). Residence time of approx. 2-4 hours. Flow must be controlled to keep outlet oxygen concentration at 2-3 mg / L or nitrate at approx. 30 mg / L. If oxygen is completely depleted extensive production of H₂S will take place, which is both toxic and bad smelling. Stable environmental conditions and correct C/N is important for operation.



Resulting production of sludge is quite high, and the unit has to be back-washed, typically once a week. A tank of approx. 10 m³ seems to handle a system for 200 kg feeding a day. Too high outlet conc. of nitrite is very often a problem, small units are recommended

Picture 12. Denitrification tower, with pump for supply of methanole

Design of biofilters

Biological filters are based on adsorption and biodegradation of impurities in the water. Micro organism can be free living suspended in the water--activated sludge systems, or they can grow on a surface media. In municipal waste water, activated sludge systems are the most widespread. They have higher turn-over in a given volume than other forms and thereby lower costs. This technique cannot be used in fish farming, as outlet water is very low in nutrients. Due to the low loading, division time for the bacteria would be too long and they would be washed out of the system. Bacteria would not be able to form aggregates at densities high enough to permit settling and eventual return via pump to the bio-reactor. This feature is a fundamental requirement for activated sludge.

This leaves us with the fixed bed or fluidised bed technologies; bacteria and other micro-organisms attached to a substrate.



Picture 13. Bionet filter elements (150 m2/m3)

Submerged filters:

Filter material, submerged in a tank, can receive influent water in the following ways: up flow, down flow or even horizontal. All flow patterns require very little head loss. Normal lifting heights: 1-5 m. Sometimes extra oxygen must be added to ensure proper function. Hydraulic surface loading must be more than 0,3 m / min.





Figure 14. Horisontal flow

Trickling filters:

A very cost effective "cabin" construction, no tank construction needed. Even distribution of water at the top must be ensured. Hydraulic loading must not be less than 0.8 m / h. This type of filter is most efficient for exchange of gases i.e. oxygen to the water and CO₂ out of water. The "chimney" effect will normally ventilate the filter, air will move up or down, dependent on differences of temperature of air inside the filter and outside environment.



Rotating biological contactors (RBC):

The biological filter material continuously rotates, and shares the same advantages as trickling filters. But the construction of 1 m³ filter material is very expensive, so this type of filters is more suited to heavily loaded water. Many small aquaculture enterprises employed the RBC with very little success:



Picture 14. RBC

Fluidised bed filters:

The filter media is in suspension, very high specific surface areas available i.e. very compact units. low headloss but energy consumption for introduction of air. These units has been in widespread use in N. America. Normally with sand as the carrying media. Their performance as nitrifying biofilters has been efficient, but hydraulic design is largely undescribed. One needs to know the relationship between hydraulic loading, bed expansion and pressure drop, as they are critical to fluidised reactor design and vary according to characteristics of the granular medium (density, size and shape, porosity etc.) and by the density and viscosity of water. No easy rules of thumb can be given here. Design must be based on a semi-scale test, performed by the supplier of the system. New types exist with carrying media of different kinds of plastics with very large surface areas, 400-2000 m² / m³

As densities of beads are very close to 1, fluidisation is not as critical as for sand filters.



At low velocities they have been used as particle retention filters. However, as sludge accumulates, rate of nitrification declines. The result: loss in hydraulic conductivity as captured solids and "biofloc" fill the pores of the bed. It's therefore recommended operating these filters at higher superficial velocities avoiding sludge accumulation. Microscreens placed before these filters will reduce particle loading throughout the entire system.

Operated at high hydraulic loadings (i.e. air introduction) these plastic beads will slough of dead bacteria, maintaining a very "fresh" bio-film at outer surfaces. Some beads are constructed as small round pieces with an internal cross section, approx. 1 cm \times 0,5 cm. Their efficiencies have been reported to be higher per m², than traditional filters, down to 40 m² for each kg feed applied to the system. This indicates that these beads seem to work partly as a carrier-substrate for a kind of activated sludge, yielding higher weights of living biomass per m³ volume of biofilters.

Technology has to be adapted to farmed species as well:

The first part has been related to the economic characteristics of a RAS, to ensure optimal environment for a given fish or crustacean, a carefull selection of systems must be performed:

Type of	BOD	Suspended	NO ₃ -N	NO ₂ -N	NH ₃ -N	Org. N	Tot-N
biofilters	5	solids	3	2	3	-	
Sandfilter							
+ filtration	low	low	high	low	low	low	medium
÷ filtration	var	var	high	var	var	var	high
RBC							
+ filtration	low	medium	high	low	low	low	medium
+ filtration	high	high	high	var	var	high	high
Trickling Filter							
+ filtration	low	medium	hiah	low	low	medium	medium
÷ filtration	high	high	high	var	var	high	high
Trickling + Submerged filter Incl particle filtration	Low	Low	Medium	Low	low	Low	medium
Submerged F. Incl particle Filtration	Low	Low	Medium	Medium	Medium	Low	medium
Moving bed Filter. Filtration before biofilter*	Medium	Medium	High	Low	Low	Medium	high
Moving bed Filter. Filtration before and after biofilter	Low	Low	High	Low	Low	Low	high

* Microscreens are usually an integrated part of systems wth moving bed biofilters. Filter cloth openings of 60-80 μ m before biological filter and 20 μ m after.

Biofilters combined with mechanical filtration are the essential parts of any water purification system. There are many variations and combinations. If there is a "var" sign in above tabel, it means fluctuations, which is not acceptable in a commercial fish farm. Low, medium and high are relative figures, but it indicates water characteristics which may be critical for a given fish species. E.g if you plan to operate a Tilapia farm, you may not worry about a high concentration of suspended solids, as long as BOD, ammonia and nitrite is low. A trout or hatchery manager will worry a lot about suspended solids and they will have to look for a combination of biological and mechanical filters to secure low turbidity.

As it can be seen from above table, all systems are designed to end up with quite high concentrations of nitrate.

Nitrate forms the main part of total nitrogen and may as well be the limiting factor for proper performance of a given fish species. It has therefore become a common component to have a denitrification unit as part of the system, thereby reducing nitrate to acceptable levels.

6. DISCUSSION

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Design of most systems today are likely to maintain a water quality which meets the needs of the farmed fish (see appendix 2 for a sketch of a typical recirculated plant). Management of the systems must be emphasised, as biofilters must be treated as a living organism.

Water quality.		
Parameter	Normal range	Toxic effect
Oxygen, O ₂	6-9 PPM	< 5 PPM
Nitrogen, Ñ,	80-100 % saturation	> 100 % saturation
Carbondioxide, CO ₂	10-20 PPM	> 20 PPM
Ammonia, NH3/NH4-N	0-5 PPM (pH dependent)	> 5 PPM
Nitrite, NO ₂ -N	0-1,5 PPM	> 1,5 PPM
NO ²	0-5 PPM	> 5 PPM
Nitrate, NO_3^2 -N	50-100 PPM	> 90 PPM
NO ₃	200-400 PPM	> 400 PPM
BOD	5-20 PPM	N.A
Alkalinity	2-5 mekv./L	< 1 mekv./L
pH	6,5-8,5	< 6 and > 8,5
Temperature	According to species	

Toxic levels are not fixed, there are differences between species and some chemicals have a synergistic effect. The above figures are only guidelines, to indicate range of acceptable environment. These figures must be measured regularly. This is a very big difference between closed systems and open flow through systems. In the latter case, fish farmers have not been accustomed to water quality monitoring. Fixed schedules and routines must be implemented, as purification systems must be running at optimum conditions at all times.

Design of water treatment in a closed system must emphasise:

- Particle filtration.
- Aerobic conditions.
- How to avoid accumulation of sludge.

Degradation of organic material and nitrification are the main processes performing in bioreactors, resulting in growth of bacteria:

Definition of bacterial growth. Biomass: X (g) Growth: Δ X (g) Time: Δ t (day)

Yield. Y (yield) = Δ biomass/ Δ substrate = Δ X / Δ S (g / g)

Growth rate.

 μ (growth rate) = $\Delta X / X \times \Delta t$ (g/g×day).

table 1:

	Heterotrophe bacteria	Nitrification bacteria
u. max. 20 °C	4-8	0.4-0.6
Ŷ	0.5-0.7	0.15-0.2
 1 10 1 1 .		

Y can be used for calculation of sludge production

Substrate for heterotrophe bacteria is organic material (BOD) and ammonium for nitrifying bacteria. As can be seen, from above table 1, growth in relation to substrate is much higher for heterotrophe bacteria.

Biofilters must be designed to operate as nitrification filters

At less than 5 PPM BOD, heterotrophe bacteria will be limited in growth by lack of substrate, conditions will be in favour of nitrification i.e. nitrosomonas and nitrobacter will have a higher growth rate. This can only be achieved by a combination of feed management, sizing of surface area and microfiltration before biofilters.

From biochemical literature it can be concluded, that conc. BOD / conc. O_2 , must be less than 5, i.e. if inlet of BOD is 20 mg/L, then O_2 , must be monitored at a conc. higher than 4 mg / L.

If conc. $NH_4 / conc. O_2$, is less than 0,3-0,4-- ammonia will be limiting substrate and reaction kinetics has been proved to be close to 1'st-order reaction. I.e. oxidation proportional with conc. of ammonia, this is the case in most recirculated systems as ammonia concentration is quite low. In practical terms it means that at proper sizing of biofilters, diurnal fluctuations in ammonia production can be absorbed, without significant increase of ammonia in the system. Normally a fixed value for removal kinetics are used for sizing of biofilters i.e. 0,7 g NH_4 / m^2 , Oorder kinetics. But as has been discussed, this is only true within some limits.

Design must reflect these relations--characterised by smaller units and constant hydraulic loading. The biofilm has to be kept in a constant state of fresh growth, nutrients and gas exchange is performed through convection and diffusion into the layer of micro-organism constituting the biofilm, diffusion is only effective in layers of 0,5 - 1,0 mm.



Figure 17. Condition of bio film

In fluidised bed filters the "fresh" condition is maintained continuously. In submerged filters proper back-wash procedures must be implemented. Sludge will be accumulated to some extent in submerged filters and can be washed out of the system during back-washing. In fluidised filters, sloughed of bacteria will accumulate in the system.

A combination of small units of submerged filters and trickling filters (2/3 of surface areas in submerged and 1/3 in trickling filters) seems to be a very reliable combination.

Fluidised biofilters are efficient in relation to biofilm kinetics, but are more sensitive in respect of intermittent break down on energy supply etc. Combinations of fluidised, submerged and trickling filters might combine advantages related to each filter type, but has yet to be proved! In any case efficient removal and procedures to avoid organic loading is imperative.

Carbon dioxide

Production of CO₂ is related to oxygen consumption in fish and degradation of organic material in biological filters. At respiratorial quotient (RQ) of 1 (production of CO₂ / O₂ consumption), approx. 700 g O₂ (consumption from 1 kg feed) will be converted to 1000 g CO₂. If not "stripped" out of the system pH will decrease. :

$$H_2CO_3 \Leftrightarrow CO_2 + H_2O$$

The carbon dioxide content of natural waters is low and often almost nil. CO_2 is extremely soluble in water and diffusion is rapid, compared to oxygen. Most of the CO_2 enters into H_2CO_3 (carbonic acid) or is found in the form of carbonates or bicarbonates.

In a recirculated system, pH would drop very fast if CO_2 concentration were allowed to increase. Systems to "strip" CO_2 must be part of design:

- Aeration in trickling filters, as these filters normally are designed as nitrification filters, gas exchange will be efficient.
- Introduction of air, CO₂ content in atmospheric. air is only 0,03 %. In practical terms approx. 80 100 m³ of air has to be introduced for each kg feed.

Toxic effects of carbon dioxide are both related to lowering of pH and on respiration, as it will influence the shape of oxygen dissociation curve of the blood, thereby reducing oxygen uptake. Buildings must be ventilated to keep room CO₂ at low levels.

Control of pH

Production of acids:

1) Carbonic acid, "stripping" as mentioned above.

2) Nitrification, $NH_4 + 2O_2 \iff NO_3 + 2H^+ + 2H_2O_3$

For each kg feed at (FC = 1), 50 g N has to be oxidised, yielding 7,1 equivalents of acids.

Denitrification will remove acid:

 $4NO_3 + 4H^+ \iff 5O_2 + 2H_2O$

50 % of produced acids will be removed, through denitrification of equivalent amount of nitrogen.

Consumption of buffer is dependent on how extensive denitrification will take place and the alkalinity of water.

Neutralisation by Ca(OH)₂:

From the above calculations it can be seen that for complete neutralisation, max. 284 g base must be applied to the system (for each kg feed) to maintain pH. This is never the case as denitrification will take place and buffer will be added through water exchange. In average 25 % of above is added.

Neutralisation by NaHCO₃:

In terms of weight, twice as much must be applied, compared to calcium hydroxide. Bicarbonate is a weaker base, but safer to use. A saturated solution has a pH of 9,5, versus calcium hydroxide that has a corresponding pH level of 11,5. In smaller systems bicarbonate is recommended. Hydroxides are used in bigger systems as costs are much lower.

Water flow and oxygenation

The circulating water has two functions:

- removal of waste from fish tanks.
- transport of oxygen.

Oxygen:

Transport of oxygen to tanks is normally the sizing factor for water flow. Oxygen consumption is not a fixed figure, but are fluctuating according to feeding activity. A level of 400 mg O_2 / kg fish × hour, is normally used as upper limit for consumption.

Example: 1.000 kg fish O_2 consumption: 400 mg × 1.000 kg = 400 g O_2 / h Inlet water, super saturated: 12 mg/l. Outlet water, must not be lower than 7 mg/l.

Water flow: 400 g/h / $(12 \div 7) = 80 \text{ m}^3 / \text{h}$

Stocking densities in intensive systems are normally within 50 - 100 kg fish /m³. Sizing of water flow based on slightly super saturated water, with a resulting residence time down to 7 -8 min. This is more than enough time to keep waste products at an acceptable low level. The next limiting factor is probably BOD, as it serves as substrate for bacterial growth, which should be limited to biofilters.

BOD:

If feeding level is 3 % / d, of standing stock: 1.000 kg \times 0,03 = 30 kg feed / d

 $30 \text{ kg} \times 0,338 \text{ kg} = 10,1 \text{ kg}$ BOD.

At given flow of 80 m³/ h: \triangle BOD = 10.100 g / 80 × 24 = 5,3 mg / 1.

To avoid bacterial growth in the tanks, residence time should not be more than 30 min., as some micro-organism has a division time of even less, and they would thereby not be flushed out of the tanks, but could in theory multiply in tanks, causing a "stressing" environment for the fish.

As can be seen from the above calculations, sizing of change of water in the tanks, must be based on acceptable oxygen levels. A residence time of less than 30 min will keep negative effects of wastes at acceptable low limits.

Water flow:

Two pipelines to the tanks would be the best construction, one line with super saturated water and one with normal atm. saturated water. This would secure change of water and oxygen supply, independent of stocking density in each tank. In one line systems, super saturation will appear in tanks at low stocking densities, as regulation has to be based on highest biomass.

Example:

In a 10 m³ tank, stocking density 100 kg / m³, total max. oxygen consumption 400 g / h. Total max. water flow 30 m³/ h (residence time approx. 20 min.), min O_2 outlet conc. 7 mg / l :

Supply of oxygen:

Saturated water: (100 % saturation at i.e. 20 °C = 9 mg / 1) 50 % of water flow = 15 m³ / h, (9 mg / 1 - 7 mg / 1) = 2 × 15 = 30 g / h. Super saturated water: (350 % saturation at 20 °C = 32 mg / 1) 50 % of water flow = 15 m³ / h, (32 mg / 1 - 7 mg / 1) = 25× 15 = 375 g / h.

Total oxygen supply:-----: 400 g / h

Oxygen consumption in biological filters for nitrification and degradation of organic material is at the same level as consumption in tanks. Oxygen supply is given by construction--- fluidised beds and trickling filters are extensively aerated, submerged filters are normally run as the first step without additional oxygen supply, as inlet should be outlet conc. from fish tanks, approx. 7 mg / l.

Oxygenation systems

Oxygen supply must be secured at all times; both the main supply and emergency systems. A lack of oxygen supply is lethal within a few minutes at these high stocking densities. The oxygen content of air is about 20 times that of water saturated with air. To increase transport capacity, water must be super saturated. Pure oxygen is employed, increasing partial pressure from 0,21 atm. (21 % oxygen in atm. air) to 1,0 atm.. Solubility is proportional with pressure i.e. at 20°C, 100 % saturation corresponds to 9 mg / 1 (see table). At pure oxygen atm., saturation level will be : $5 \times 9 = 45$ mg / 1. Due to five times increase in partial pressure. Pressure can be added in the oxygenation system as well.

One of the most widespread systems in use is the oxygen cone:



Picture 15. Oxygen cones in an eel farm

Principle: Water is pumped through the cone from the top, pure oxygen is added. Only pure oxygen must be introduced, as nitrogen, which constitutes 78 % of atm. air, at super saturation of only 3 - 5 % will cause severe damages to the fish.



Figure 18. The oxygen cone

Oxygen is trapped inside the cone, as velocity (V) at inlet is higher than upflow velocity of oxygen bubbles. Water velocity at the bottom is lower than up flow velocity of bubbles, thereby trapping the oxygen. Oxygen cones are operated at a pressure 1 atm above normal pressure. A properly designed cone operates at 80-85 % efficiency of added oxygen. Efficiency is very dependent of water flow, supply of oxygen and added pressure. Pure oxygen is usually quite expensive and systems for oxygen enrichment are therefore sized for optimum efficiency of supplied oxygen. In the cone system optimum efficiency of added oxygen is at a level

corresponding to approx. 40 % of saturation at given pressure. At mentioned above figures of 9 mg / 1 (atm. saturation at 20,4 C see appendix 3), saturation level at 100% oxygen can be calculated as solubility of a gas is proportional to partial pressure, which is five times higher, using pure oxygen: $5 \times 9 = 45$, pressure in cone 2 atm.: $2 \times 45 = 90$ mg/l, outlet conc. from cone: $0,4 \times 90 = 36$ mg/L. The cone is a flexible system, oxygen conc. can be monitored in the tanks and regulated accordingly by adding or reducing oxygen supply to the cone. Operating costs of this system is related to the energy consumption of pumping, 3-4 kW for each kg oxygen dissolved and the price of oxygen. An emergency system must be mounted in each tank, adding oxygen directly through a diffuser system. Back-up oxygen must be independent of electrical supply.

APPENDIX 1A: SLUDGE CONCENTRATION

Resulting sludge production from filtration in fish farms are in many cases a storage and economic problem. Below the figure is a case study from a smolt farm in Norway, done by Rogaland research. Four microscreens Hydrotech HDF1607, receives outlet water from fish tanks. Total Water flow 2.076 m3/h, first filtration through 90 micron filter cloth, efficiency of filtration on particulate dry matter(D.M), 36 - 54 % - but based on very low inlet concentrations 1-3 mg / L. Sludge water from filters, flow by gravitation, to one microscreen model HDF1602, filtration at 80 microns. Concentration of D.M in sludge water approx. 1.000 mg / L, flow of sludge water 240 - 2.400 L/ h., depending on dis- or continuous running of filters.

Efficiency of model HDF1602, 90 - 99 % on D.M, concentration of sludge approx.. 4 times, i.e. 4.000 mg D.M / L. 60 - 600 L/ h, is pumped to concentration tank. Dimensions of tank, width: 2,0 m, volume: 5,5 m3, hydraulic loading: 0,02 - 0,18 m/h. Residence time: 9 h to 3,8 days.

Sludge were stabilised in a small tank, adding 8 kg CaO / 500 L sludge, increasing pH to 11 - 12. At this pH all pathogens are destroyed. Storage tank for approx. 1 months production. Production 0,5 L sludge for each kg feed or 50-70 g D.M, corresponding to 10-14 % D.M

Above installation were monitored in a situation with very low loading on first filtration, efficiency is higher at higher loadings. Concentration of D.M is increased four times after sec. filtration, before final sedimentation. Cost / benefit must be evaluated, if increase of size of settling tank is more economic, there might be no benefit of sec. filtration.



APPENDIX 1C: SLUDGE CONCENTRATION, BELTFILTER

Further concentration of sludge from microfilters and also back wash water from biofilters in some RAS, is possible by adding polymers followed by draining on beltfilters.

By adding this technology TSS in resulting sludge, is increased from 0,05-0,1 % up to 8 %-12 %.

The filtrated water is returned to the system, usually through a de-nitrification unit, which will reduce an otherwise unacceptable high level of nitrate. In some systems without de-nitrification units, water is piped back in front of biological filters.



First step is effluent treatment by drumfilter or discfilter



A mixer tank and a beltfilter for sludge concentration

APPENDIX 2: FLOW DIAGRAM, RECIRCULATED FARM

SKETCH FOR RECIRCULATED PLANT



Sludge for sludge tank





APPENDIX 3A: SATURATION OF OXYGEN IN FRESH WATER

Assumptions: pressure = 760 mm salinity = 0,0 ppt

T. T.

Example: T = 15,1C° DO = 10,05

Temperature

Temperature, degrees Celsius										
Whole numbers	Tenth of	degrees	Celsius							
	.0	.1	.2	.3	.4	.5	.6	.7	.8	.9
0	14.60	14.56	14 52	14.48	14.44	14.40	14.36	14.32	14.28	14.24
1	14.20	14.16	14.12	14 08	14.04	14.00	13.97	13.93	13.89	13.85
2	13.81	13.78	13.74	13.70	13.66	13.63	13.59	13.55	13.52	13.48
3	13.45	13.41	13.37	13.34	13.30	13.27	13.23	13.20	13.16	13.13
4	13.09	13.06	13.03	12.99	12.96	12.92	12.89	12.86	12.82	12.79
5	12.76	12.72	12.69	12.66	12.63	12.59	12.56	12.53	12.50	12.47
6	12.44	12.40	12.37	12.34	12.31	12.28	12.25	12.22	12.19	12.16
7	12.13	12.10	12.07	12.04	12.01	11.98	11.95	11.92	11.89	11.86
8	11.83	11.80	11.77	11.75	11.72	11.69	11.66	11.63	11.60	11.58
9	11.55	11.52	11.49	11.47	11.44	11.41	11.38	11.36	11.33	11.30
10	11.28	11.25	11.22	11.20	11.17	11.15	11.12	11.09	11.07	11.04
11	11.02	10.99	10.97	10.94	10.92	10.89	10.86	10.84	10.82	10.79
12	10.77	10.74	10.72	10.69	10.67	10.64	10.62	10.60	10.57	10.55
13	10.53	10.50	10.48	10.46	10.43	13.41	10.39	10.36	10.34	10.32
14	10.29	10.27	10.25	10.23	10.20	10.18	10.16	10.14	10.12	10.09
15	10.07	10.05	10.03	10.01	9.99	9.96	9.94	9.92	9.90	9.88
16	9.86	9.84	9.82	9.79	9.77	9.75	9.73	9.71	9.69	9.67
17	9.65	9.63	9.61	9.59	9.57	9.55	9.53	9.51	9.49	9.47
18	9.45	9.43	9.41	9.39	9.38	9.36	9.34	9.32	9.30	9.28
19	9.26	9.24	8.22	9.21	9.19	9.17	9.15	8.13	9.11	9.09
20	9.08	9.06	9.04	9.02	9.00	8.99	8.97	8.95	8.93	8.92
21	8.90	8.88	8.86	8.85	8.83	8.81	8.79	8.78	8.76	8.74
22	8.73	8.71	8.69	8.68	8.66	8.64	8.63	8.61	8.59	8.58
23	8.56	3.54	8.53	8.51	8.50	8.48	8.46	8.45	8.43	8.42
24	8.40	8.38	8.37	8.35	8.34	8.32	8.31	8.29	8.27	8.26
25	8.24	8.23	8.21	8.20	8.18	8.17	8.15	8.14	8.12	8.11
26	8.09	8.08	8.06	8.05	8.04	8.02	8.01	7.99	7.98	7.96
27	7.95	7.93	7.92	7.91	7.89	7.88	7.86	7.85	7.84	7.82
28	7.81	7.79	7.78	1.11	1.15	7.74	7.73	7.71	7.70	7.68
29	7.67	7.66	7.64	7.63	7.62	7.60	7.59	7.58	7.57	7.55
30	7.54	7.53	7.51	7.50	7.49	7.47	7.46	7.45	7.44	7.42
31	7.41	7.40	7.39	7.37	7.30	7.35	7.34	7.32	7.31	7.30
32	7.29	1.Z1 7.45	7.20	7.20	7.24	7.23	7.21	7.20	7.19	7.18
33	7.17	7.15	7.14	7.13	7.12	7.11	7.10	7.08	7.07	7.06
34	7.05	7.04	7.03	7.01	7.00	6.99	0.98	6.97	0.90	6.95
30	0.93	0.92	0.91	6.90	0.09	0.00	0.07	0.00	0.00	0.03
30	0.02	0.01	0.00	0.79	0.70	0.77	0.70	0.75	0.74	0.73
37	0.72	0.71	0.70	0.08	0.07	0.00	0.00	0.04	0.03	0.02
38	0.01	0.00	0.09	0C.0	0.07	0.00	0.00	0.04	0.03	0.52
39	0.01	0.00	0.49	0.40 6 20	0.47	0.40	0.40	0.44	0.43	6 22
40	0.41	0.40	0.39	0.38	0.37	0.30	0.30	0.34	0.33	0,32

APPENDIX 3B: SATURATION OF OXYGEN IN SEA WATER

Assumptions: pressure = 760,0 mm

Salinity

	Salinity parts per thousand									
Temp (C)	0	5	10	15	20	25	30	35	40	45
0	14,60	14,11	13,64	13,18	12,74	12,31	11,90	11,50	11,11	10,74
1	14,20	13,73	13,27	12,83	12,40	11,98	11,58	11,20	10,83	10,46
2	13,81	13,36	12,91	12,49	12,07	11,67	11,29	10,91	10,55	10,20
3	13,45	13,00	12,58	12,16	11,76	11,38	11,00	10,64	10,29	9,95
4	13,09	12,67	12,25	11,85	11,47	11,09	10,73	10,38	10,04	9,71
5	12,76	12,34	11,94	11,56	11,18	10,82	10,47	10,13	9,80	9,48
6	12,44	12,04	11,65	11,27	10,91	10,56	10,22	9,89	9,57	9,27
7	12,13	11,74	11.37	11.00	10.65	10.31	9.98	9.66	9.35	9.06
8	11.83	11,46	11.09	10,74	10.40	10.07	9.75	9,44	9,14	8.85
9	11,55	11,19	13.83	10.49	10.16	9,94	9,53	9.23	8.94	8.66
10	11.28	10.92	10,58	10.25	9,93	9.62	9.32	9.03	8.75	8,47
11	11.02	10.67	10.34	10.02	9,71	9.41	9.12	8.83	8.56	8.30
12	10,77	10,43	13,11	9.80	9.50	9.21	8.92	8.65	8.38	8.12
13	10.53	10.20	9.89	9,59	9.30	9.01	8.74	8.47	8.21	7.96
14	10,29	9.98	9.68	9.38	9.10	8.82	8.55	8.30	8,04	7.80
15	10,07	9,77	9,47	9.19	8,91	8.64	8.38	8,13	7.88	7.65
16	9.86	9.56	9.28	9.00	8.73	8,47	8.21	7.97	7,73	7.50
17	9.65	9.36	9.09	8.82	8.55	8.30	8.05	7.81	7.58	7.36
18	9,45	9,17	8.90	8,64	8.39	8,14	7.90	7.66	7,44	7.22
19	9.26	8.99	8.73	8,47	8.22	7.98	7,75	7.52	7.30	7,09
23	9.08	8.81	8.56	8.31	8.07	7.83	7.60	7.38	7,17	6.96
21	8.90	8,64	3.39	8.15	7,91	7.69	7,46	7,25	7.04	6.84
22	8.73	8,48	8.23	8.00	7,77	7.54	7,33	7.12	6.91	6.72
23	8.56	8.32	8.08	7.85	7.63	7.41	7.20	6.99	6.79	6.60
24	8.40	8.16	7.93	7,71	7,49	7.28	7.07	6.87	6.68	6.49
25	8,24	8,01	7,79	7.57	7.36	7,15	6.95	6.75	6.56	6.38
26	8.09	7.87	7.65	7,44	7.23	7.03	6.83	6.64	6.46	6.28
27	7.95	7,73	7.51	7.31	7,10	6.91	6.72	6.53	6,35	6.17
28	7.81	7,59	7.38	7.18	6.98	6.79	6,61	6.42	6.25	6.08
29	7.67	7,46	7.26	7.06	6.87	6.68	6.50	6.32	6.15	5.98
30	7,54	7,33	7.14	6.94	6.75	6.57	6.39	6.22	6.05	5.89
31	7,41	7.21	7.02	0.83	6.65	6,47	6.29	6.12	5.96	5.80
3Z	7.29	7,09	6.90	6.72	6.54	6.36	6.19	6.03	5.87	5.71
33	7.17	6.98	6.79	0.01	6,44	6.20	6,10	5,94	5.78	5.63
34	7,05	0.00	0.00	0.01	0,33	6,17	5.01	5.05 5.76	5.09	5.34
30	0.93	0.70	0.00	0.40	0,24	6.07 5.09	5.92	5.70	0.01 5.52	5.40
30	0.02	0.00 6 E 4	0.47	0.31	0.14	5.90	5.03 5.74	5.00 5.50	5.55	5,39
37	0.72	0.04 6.44	0.37 6.29	0.21	0.00	5.09 5.01	5,74	5.59	5,45 5 27	5.01
30	6.51	0,44 6 24	0.20 6 1 9	6.02	5.90	5.01	5.00	5.01	5.37	5.24
40	6.41	6 25	6 NQ	5.03	5.07	5.72	5.50	5 36	5.30	5,10

Example: T= 4 C and S = 35 ppt, DO =10,38mg/L