Mass mortality in RAS – Solved?

The intention of this document is to try to explain why the industry have experienced a series of mass mortalities in Recirculated Aquaculture Systems (RAS) in the last years. The mortalities are likely linked to the release of Hydrogen Sulphide (H_2S). This document explains a simple, inexpensive and practical way to mitigate the release of Hydrogen Sulphide and mass mortality.

Pisco group has, with input from AquaCircle, the Danish Industry Cluster for Aquaculture development, elaborated this document. Further research and investigations in collaboration between the industry, Universities and knowledge institutes need to be carried out to verify the details of the theory and refine the recommendations. We feel that we need to bring this information to the table now, if there is a chance that it can save fish from H_2S induced mortality in RAS systems in the near future.

The conclusion of the theory is: Always keep the Nitrate concentrations at a sufficient level!

During the last decade it has become common to use seawater in all steps of salmon production. Salt is thus used in smolt production systems, where the aim is to control fungus, to postsmolt systems where higher salinity is a prerequisite. Seawater contains more Sulphate (SO_4^{2-}) than freshwater. Elevated Sulphate concentration increase the likelyhood of increased Hydrogen Sulphide generation in the systems.

Keep enough nitrate in the system

Until recently, Nitrate (NO_3^-) have only been considered as a by-product that needed to be deluded out of the production water or removed in denitrification systems in order not to reach toxic levels. Therefore, we have never been paying attention to the nitrate levels. We however believe that it needs to be considered in connection to the mysterious mass mortalities that we have seen in RAS systems in recent years.

By studying data from various systems experiencing sudden mass mortality, we have found that many of these systems have seen a sudden drop in Nitrate 12-72 hours before the incidents.

It is our genuine belief that we have all been inattentive to the importance of Nitrate in relation to Hydrogen sulphide generation and control. The stoichemetric formula below describes a process that are central to recirculation systems. We cannot quantify the extend, but this reaction is taking place all the time in all RAS systems. Our hypothesis is that Nitrate has been the “guardian angel” of RAS systems, by keeping the Hydrogen Sulfide generation in check and avoiding toxic levels of the substance which can result in mass mortalities.

$$5H_2S + 8NO_3^- \rightarrow 5SO_4^{2-} + 4N_2 + 4H_2O + 2H^+$$

From the equation above it can be seen, that if Nitrate is present it will scavenge the toxic Hydrogen Sulphide and prevent its release into the water, which could eventually lead to fish death.

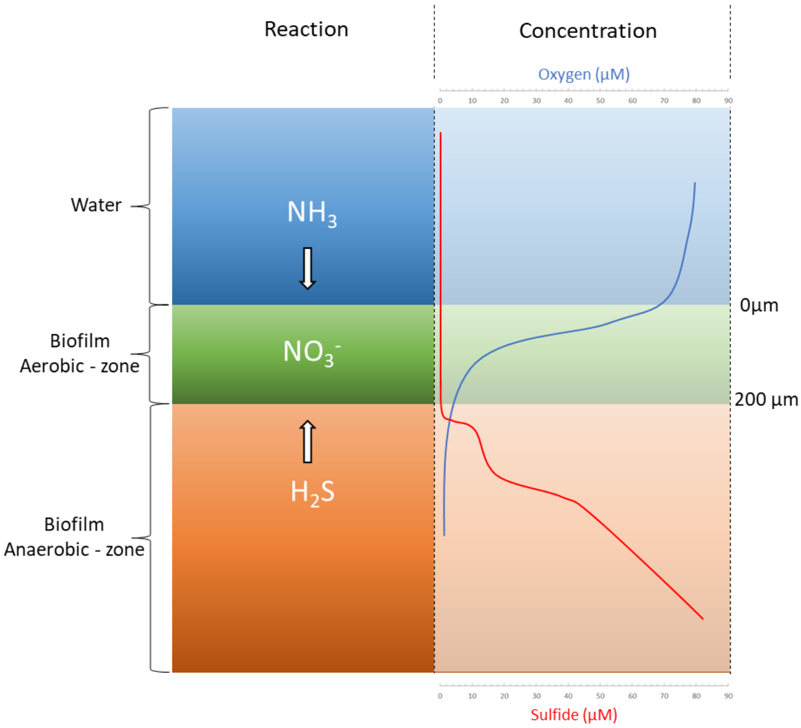
It is therefore beneficial to keep a certain amount of Nitrate in the water at all times.

This practical recommendation will be further elaborated in the next explanatory section. The following considerations about biofilm and reactions are very simplified and serves only to illustrate the principle.

Why and where is toxic hydrogen sulphide produced?

Biofilm exists on almost all surfaces in a recirculated system (not just in the biofilter). Biofilm grows and becomes thicker with age. The biofilm can simplified be viewed as two layers: An aerobic top layer (oxygen present) of approximately 200 µm and an anerobic bottom layer (oxygen depleted).

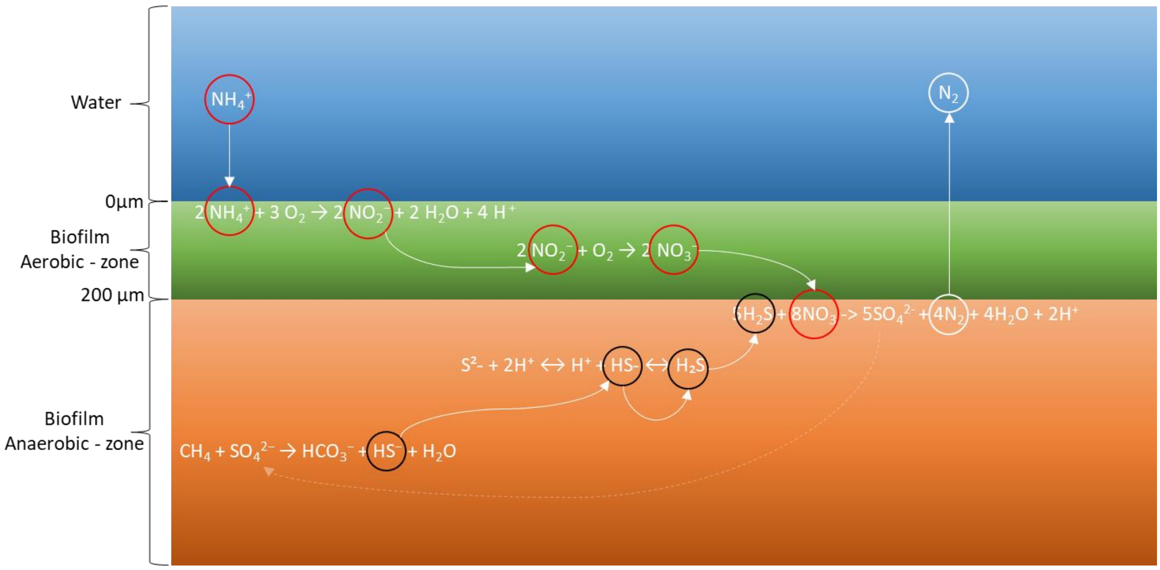
The top layer plays an active role in RAS where it converts the toxic Ammonia (NH_3) and Ammonium (NH_4^+) to harmless Nitrate through a bacterial process called nitrification. This process consumes oxygen.



The bottom layer contains bacterias that consumes organic material and converts sulphate to toxic hydrogen sulphide. This layer contains no oxygen, and can in theory be very thick and contain large ammounts of hydrogen sulphide.

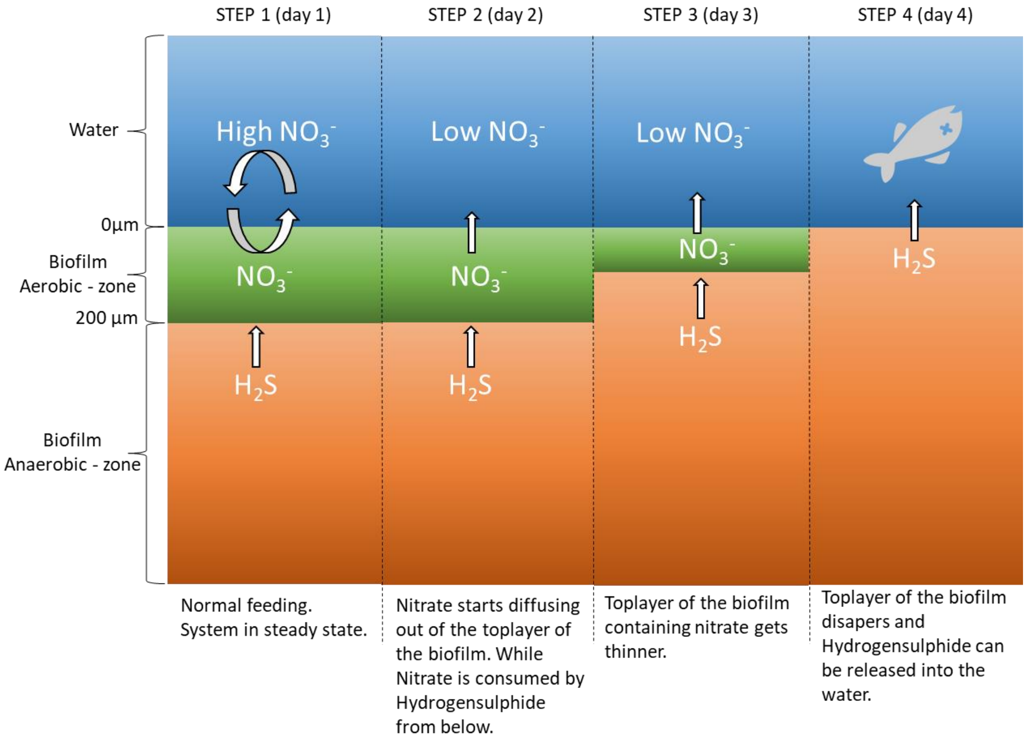
Again it must be emphazised that many other processes are taking place simultaniously in a biofilm.

The illustration below serves to further illustrate this possible pathway.



When is hydrogen sulphide released into the water?

The following example explains a common production scenario where smolts or postsmolts are starved for a number of days, before being collected by a wellboat and transported to the sea cages.



Step 1

The system is in steady state. Ammonia and Ammonium is converted to Nitrate. The concentration of Nitrate in the water is high. Hydrogen Sulphide "eats" Nitrate and coverts into harmless Sulphate.

Step 2

Feeding has stopped. The excretion of Ammonia from the fish is reduced and the bacteria convert less Ammonia into Nitrate. The concentration of Nitrate in the water decreases because of water exchange and denitrification. As the nitrification rate drops, Nitrate is depleted in the system. The Hydrogen Sulphide is still consuming the Nitrate in the bottom part of the biofilm.

Step 3

The toplayer of the biofilm is getting thinner, as the nitrifying bacteria are starved. The Hydrogen Sulphide is still "eating" the Nitrate in the bottom part of the biofilm.

Step 4

The top layer is now completely depleted of Nitrate and Hydrogen Sulphide is released into the water, with the risk of reaching toxic levels to the fish.

How can you react in time?

It is essential, always to have enough nitrate avalible in the water. Therefore you should:

- Always have Calcium nitrate $Ca(NO_3)_2$ or another Nitrate source on stock to dose into the water if the Nitrate level drops to a critical level.

- Make sure to measure Nitrate concentration at leats once a day.

- Keep the nitrate level above 40 mg/l Nitrate-(N), in order to be on the safe side.

- Reduce water exchange rate with new make up water, if you experience a sudden drop in Nitrate concentration.

There are chritical situations, where you need to pay special attention because of the risk of a sudden drop in the nitrate concentration. In these situations you should measure at least twice a day.

These situations are:

- Feeding has been stopped or reduced dramatically.
  (Remember to turn off the denitrification filter, in any, if you experience a sudden drop in Nitrate concentration)

- Large amounts of water has been changed in the system

- When restocking with fish, without having cleaned the system.
  (Remember to dose Ammonia into the biofilter during the fallow periods in order to keep the nitrifying bacteria alive)

Dosing Calcium nitrate

If the Nitrate-(N) level drops below 40 mg/l, Calcium nitrate should be dosed into the water. Calcium nitrate is a relatively inexpensive compound (approx. 10 - 20 NOK/kg). The compound can be purchased in granular- or liquid form.

For each 1,000 m^3 of water in the system, dose 5.85 kg of Calcium Nitrate to increase the concentration of Nitrate-(N) by 1 mg/l. The following equation can be used to determine the amount of Calcium Nitrate to dose into your system:

$$\mathrm{kg\ Ca(NO_3)_2\ needed} = \mathrm{Volume\ (m^3)} \cdot \frac{\mathrm{mg}}{\mathrm{l}}\ \mathrm{NO_3^-\ N\ increase} \cdot 0.00585$$

Example:

In a system with a total volume of 280 m^3, a drop down to 30 mg/L Nitrate-(N) is observed. It is therefore recommended to increase the concentration by 20 mg/l to a final level of 50 mg/l Nitrate-(N). The necessary dosing of Calcium Nitrate in granular form is thereby:

$$\mathrm{kg\ Ca(NO_3)_2\ needed} = 280\ \mathrm{m^3} \cdot 20\frac{\mathrm{mg}}{\mathrm{l}} \cdot 0.00585 = 32.8\ \mathrm{kg}$$