



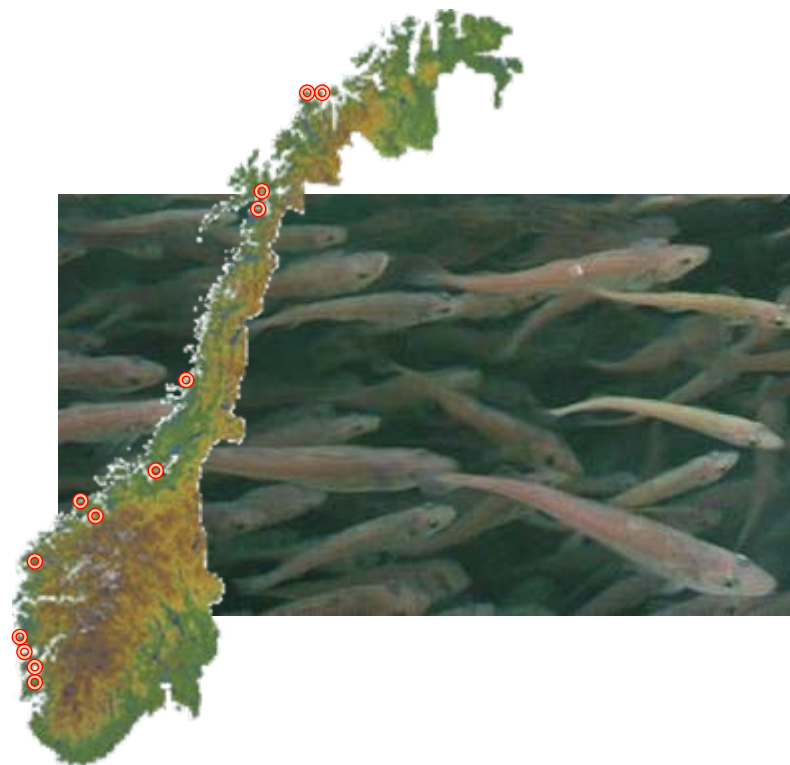
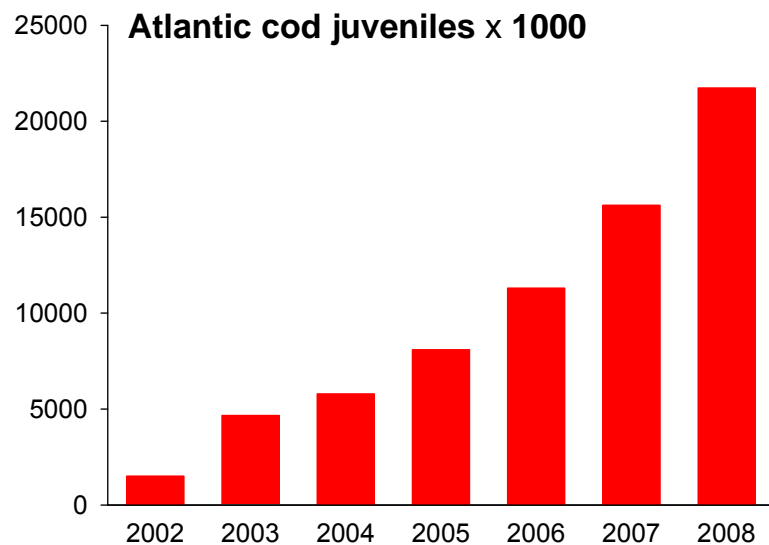
Recirculation as a microbial control strategy in intensive aquaculture of marine larvae

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Production of marine juveniles rapidly increasing, but still the bottleneck

Atlantic cod production in Norway



Similar with halibut, turbot, lobster, scallop..



The challenges

1. Bad performance

- › Survival
- › Growth
- › Development
- › First feeding



2. Lack of reproducibility

- › Same temperature
- › Same feed
- › Full sibling groups



The causes?

- Not nutrition
- Not genetics or egg quality

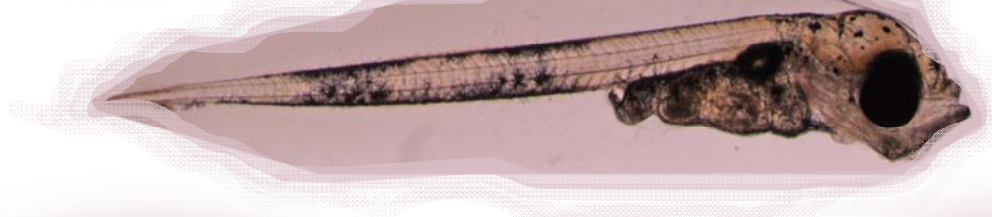
Detrimental microbial relationships?



Efforts improving juvenile production is worth while

Costly period of production
(live feed)

Sensitive larvae



Quality at later stages affected

Large growth potential

→ **A need for microbial control strategies**



General characterisation of bacteria:

Ecological r/K-theory

Carrying capacity (CC)

= Maximum sustainable biomass
 = # bacteria
 that can be maintained in a system

Determined by:

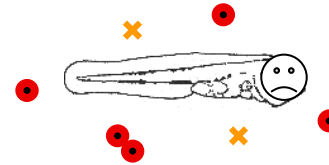
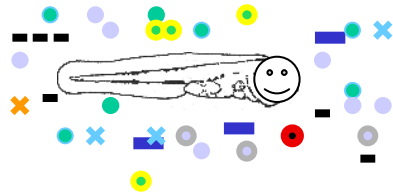
physical/density dependent restrictions
 = Supply of nutrients
 = Dissolved organic matter supply
 (heterotrophic bacteria)

Characteristics	Environment	Substrate supply per capita	Favoured ability
r-selection	Unstable or unpredictable, empty niches	High	Reproduce quickly, fast growing
K-selection	Stable or predictable, crowded	Low, close to CC	Compete for limited resources



What is a 'detrimental' fish - microbe relationship?

- › Type, composition *and* total amount

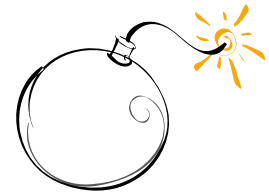


- › Larvae rely on the general immune system
- › Most diseases caused by opportunists becoming pathogenic when host's resistance is lowered by environmental stress

High share of r-selected opportunists



Unstable, low diversity microbial community





Effect of intensive fish cultivation procedures on the microbial community

Landbased fishfarm

- › Disinfection
- › Frequent perturbations
- › High and fluctuating [org. mat.]
- › High densities of fish and prey with associated microflora

**Destabilization
r-selection**

Favors proliferation of opportunistic and potentially harmful species

Selecting for the 'dodgy' guys...

Fish larvae in natural habitat

Algae, low org. mat., high redox potential, few bacteria

Stable

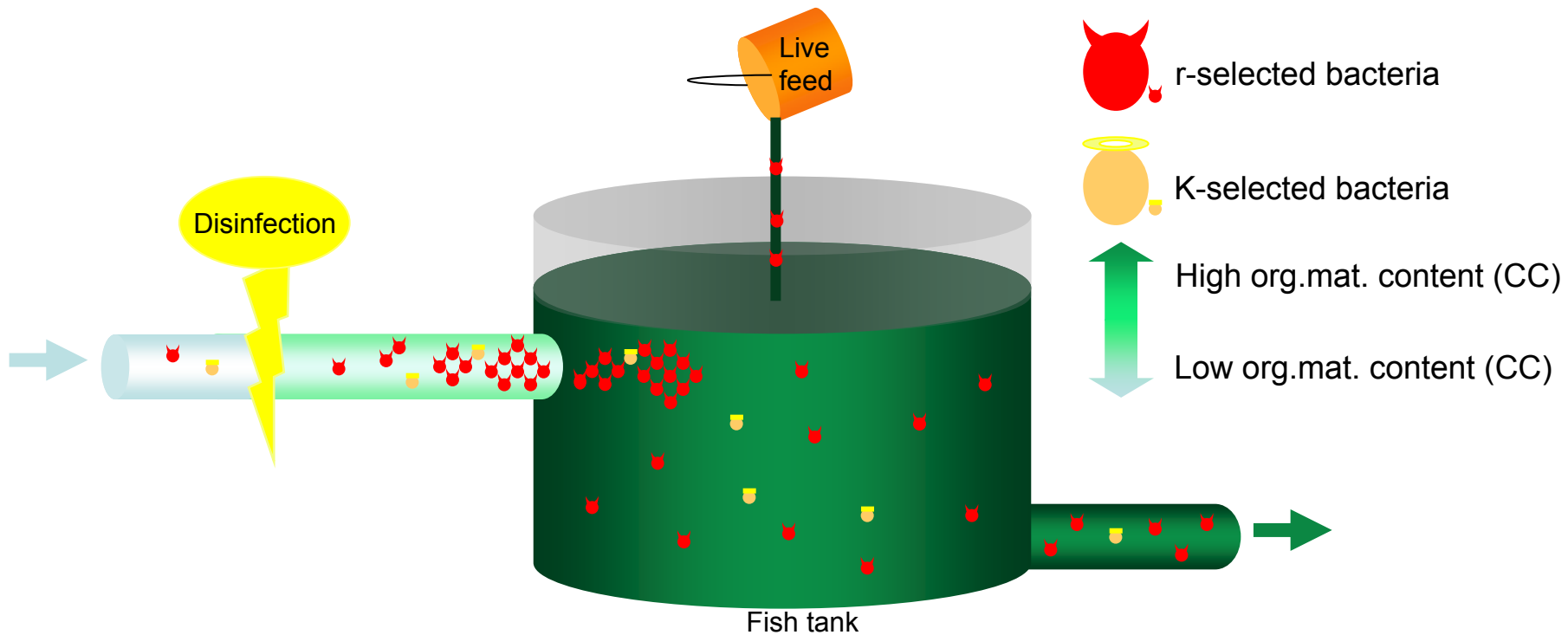
Seabed

Particles, org. mat., bacteria

Intake



Flow through





Controlled recolonization: Microbial maturation of water

- › Allow the microbial flora to stabilize at a given CC
- › Selecting for the domination of a diverse community of slower growing specialists
- › Waiting for succession and K-selection to happen

- › Shown to benefit marine fish larvae (survival, growth)

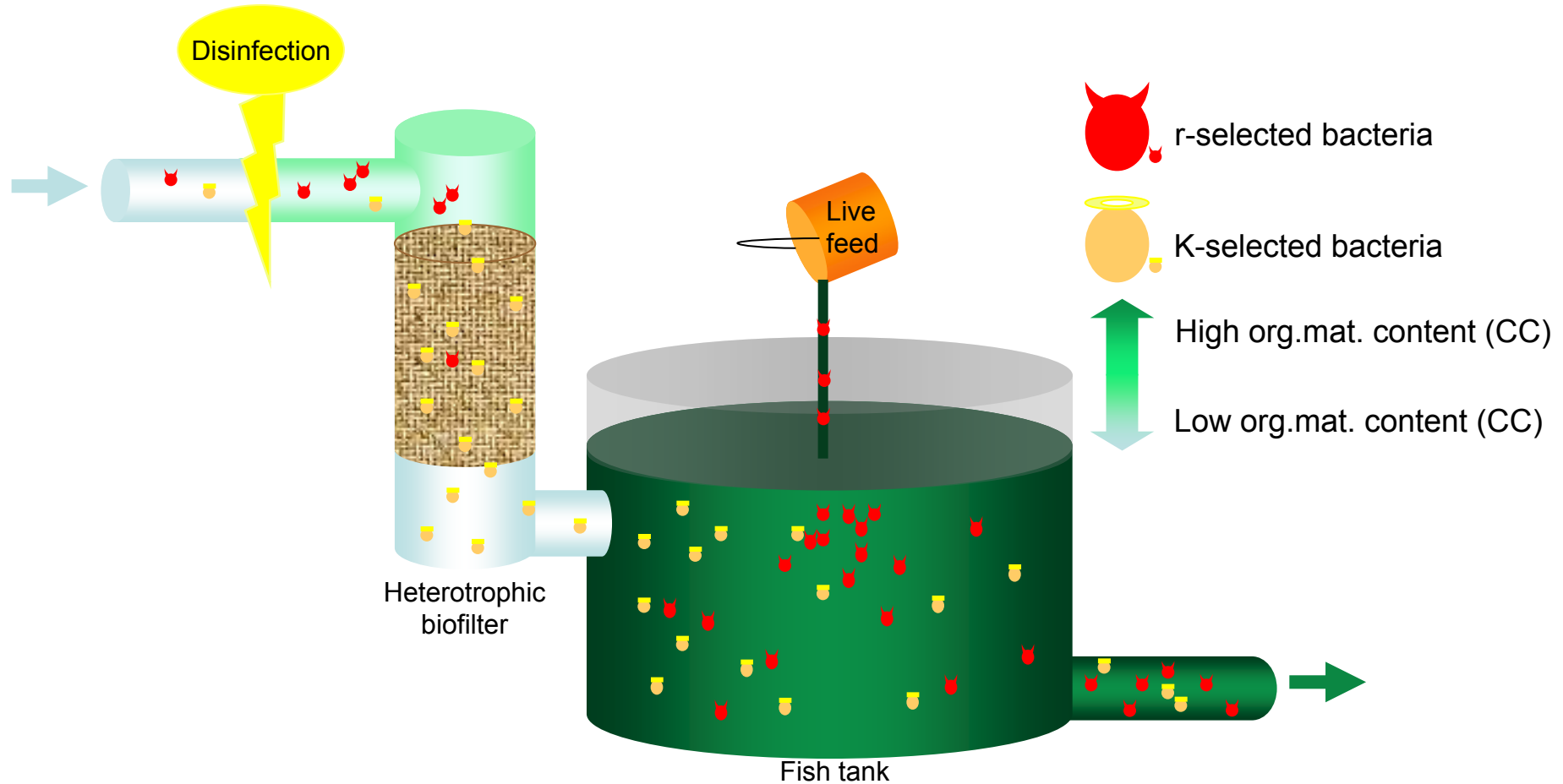
(Vadstein *et al.*, 1993, Skjermo *et al.*, 1996, Skjermo *et al.*, 1997 Salvesen *et al.*, 1999)

- › Maturing unit = biofilter = large surface area to maintain a heterotrophic biofilm influencing the intake water



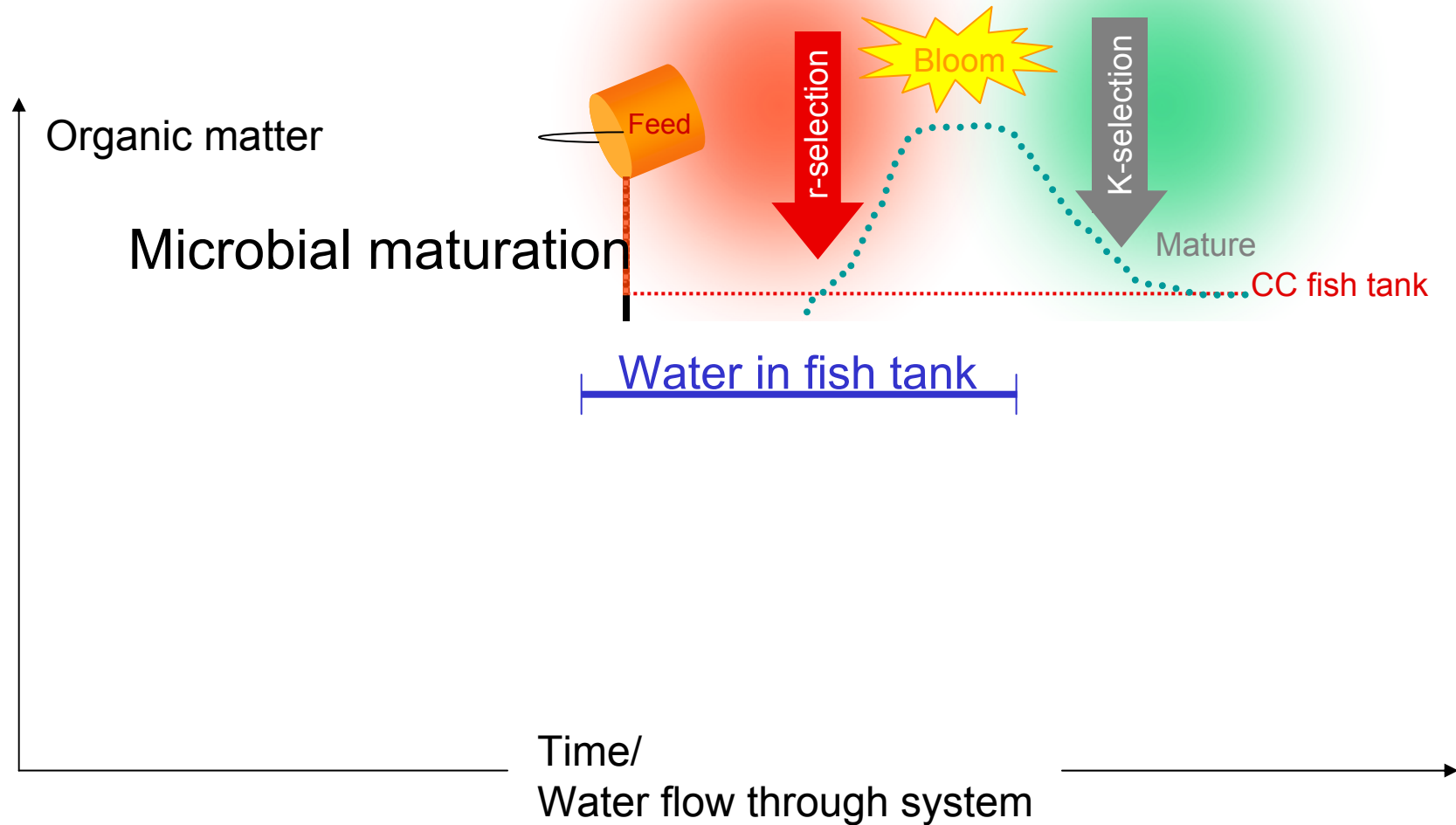
Microbial maturation, flow through

..but there is a shortage:





..a challenge to maintain the mature situation in the rearing tanks with low water exchange rates





Improved microbial maturation

To promote and maintain a stable and beneficial bacterial community

selection should be carried out at a CC similar to CC in the rearing tanks

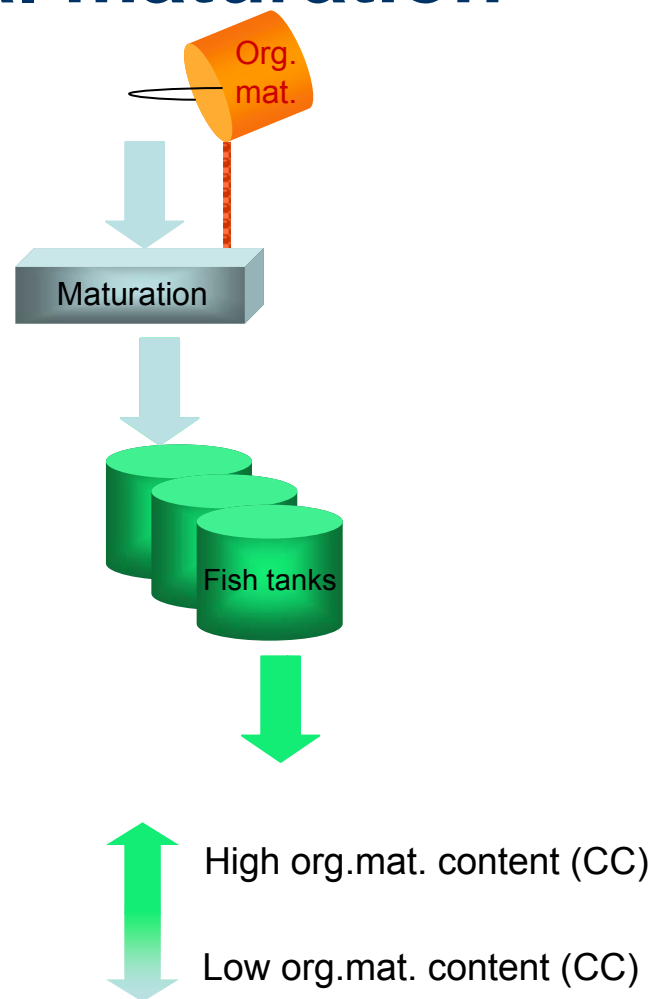
= no free niches for opportunists

1. Feed the maturing water with organic matter
2. Reuse water from the fish tanks
= organic matter source
to raise CC in the maturing unit
= biofilter



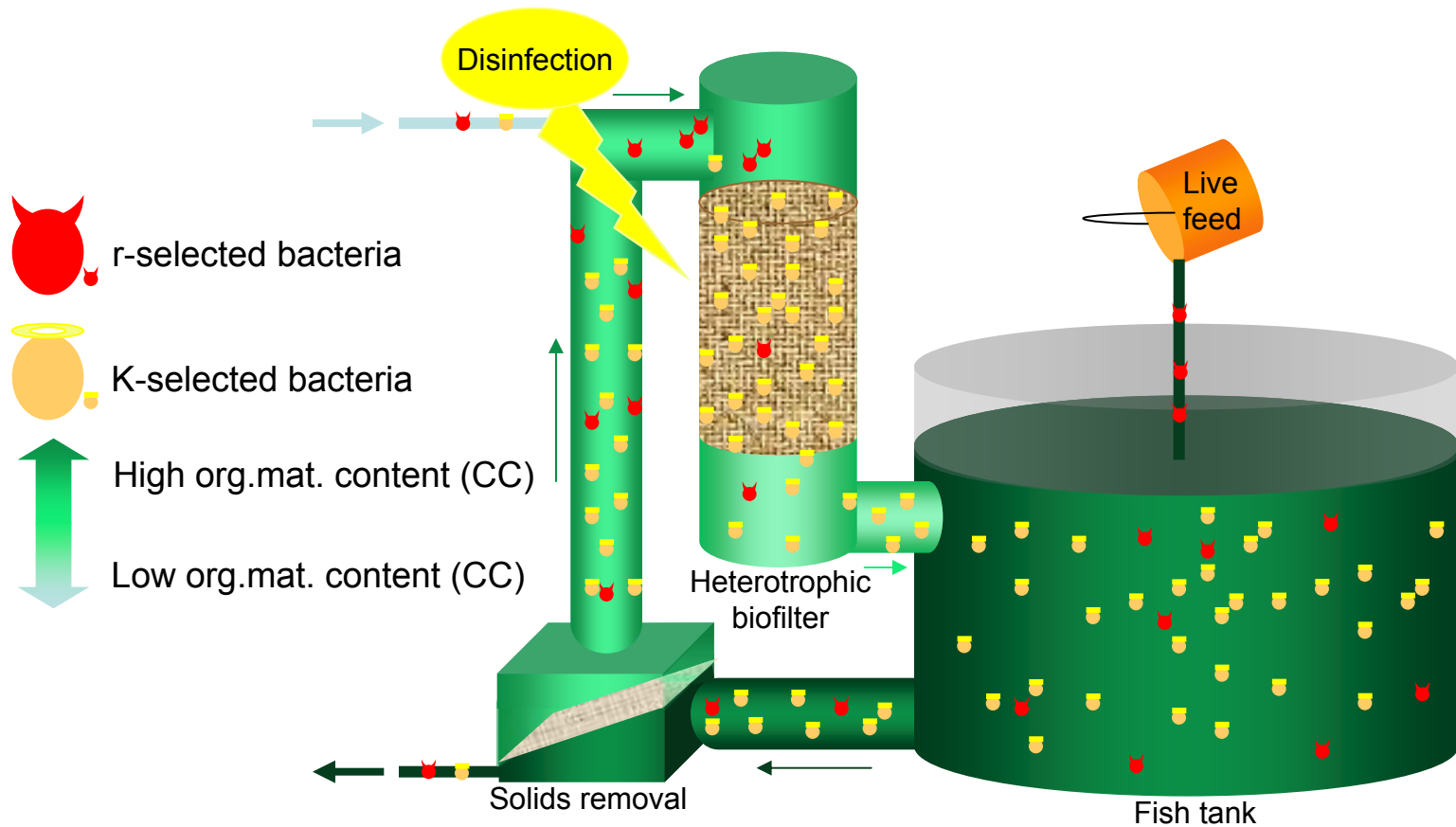
Simplest way to secure a quantitatively and qualitatively similar mix of nutrients to that found in the fish tanks

Better resource management





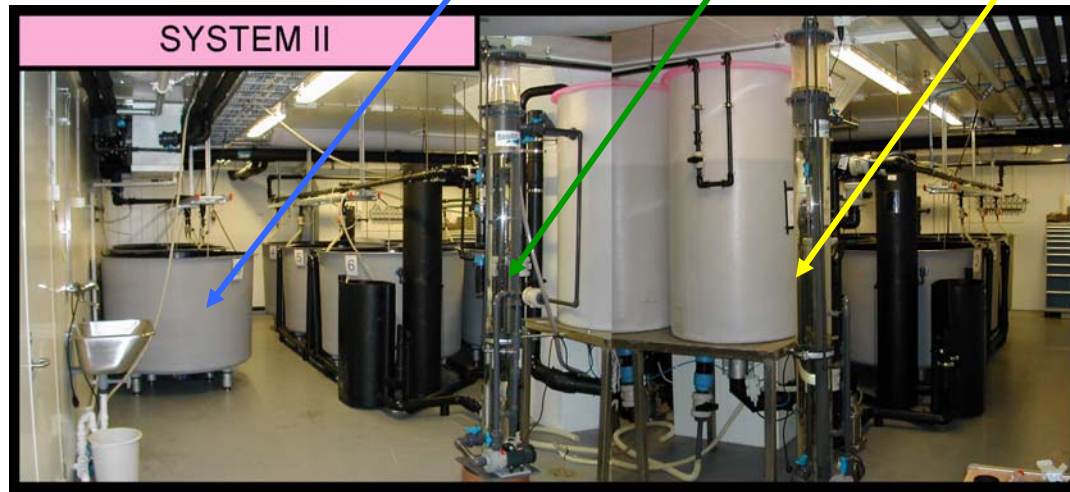
Recirculation as a microbial control strategy





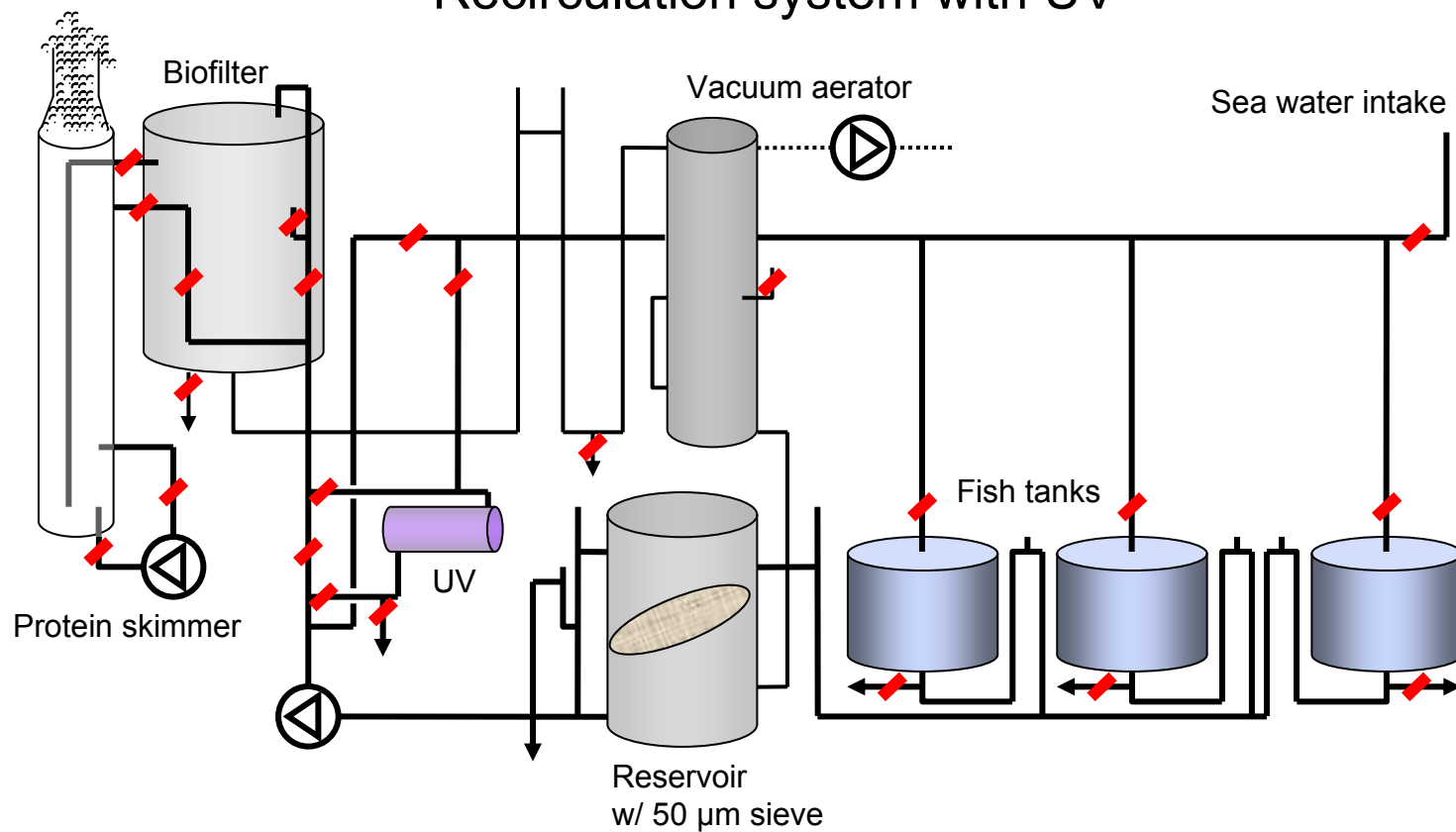
Study: RAS microbial development

- Atlantic cod (*Gadus morhua*)
- Experiment I: FTS vs. RAS w/ O₃
- Experiment II: FTS vs. RAS w/ O₃ vs. RAS w/ UV





Recirculation system with UV





Study: RAS microbial development

- **Characteristic markers for mature water:**
 - › a more stable microbial community composition over time
 - › less variability between parallel tanks
 - › higher species richness
 - › lower fraction of opportunists
- **Better performance of fish larvae?**

Result 1:



Result 2:

More mature microbial environment in RAS

- › **RAS developed and maintained a more mature, diverse and stable microbial community**
 - › Higher and more stable **species richness** and **diversity**
 - › Lower share of **opportunists**
 - › More stable microbial community **composition** over time
 - › Lower **variation** between parallel tanks
- › **Qualitative microbial shifts in FTS**
- › **Quantitative response in RAS**



Result 3:

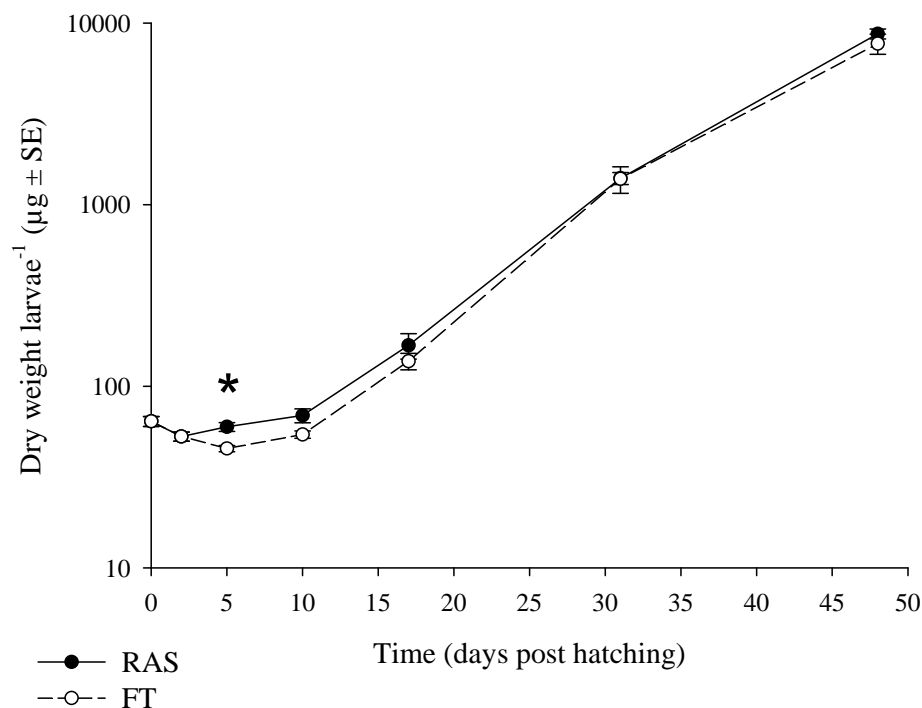
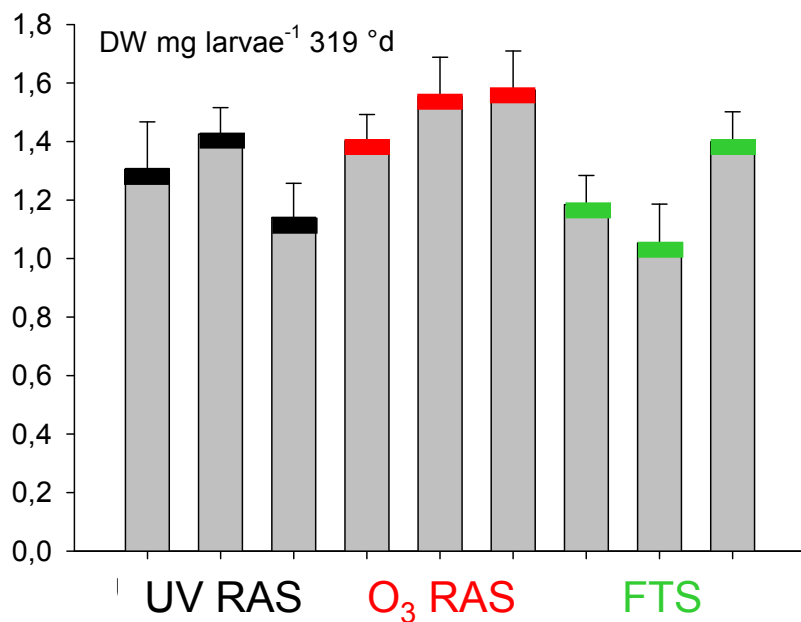
Slightly worse physiochemical environment in RAS

- More variation over time
- Higher concentrations of nitrogenous waste products
- More suspended particles
- Higher organic matter level
- Lower pH

This may be improved



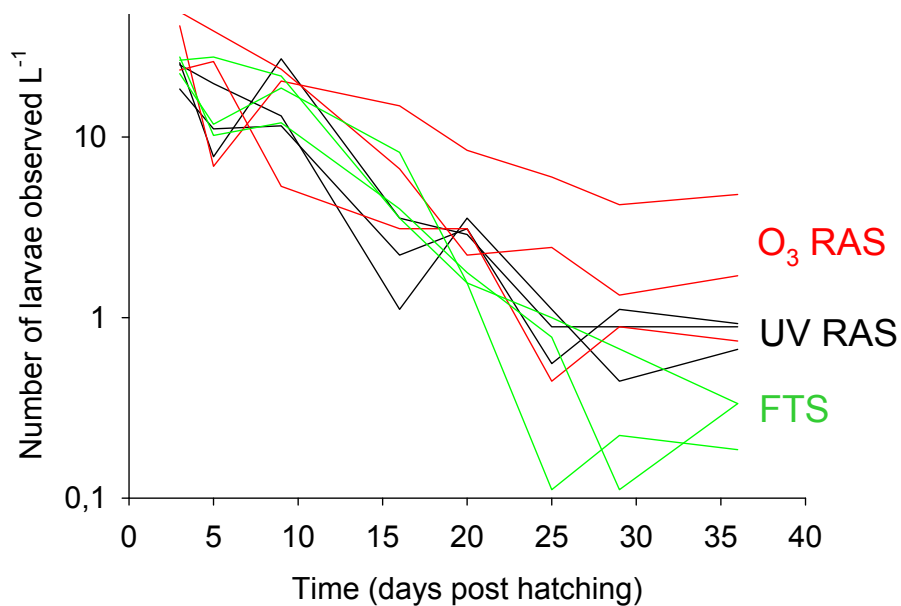
Result 4: Equal or better fish growth in RAS



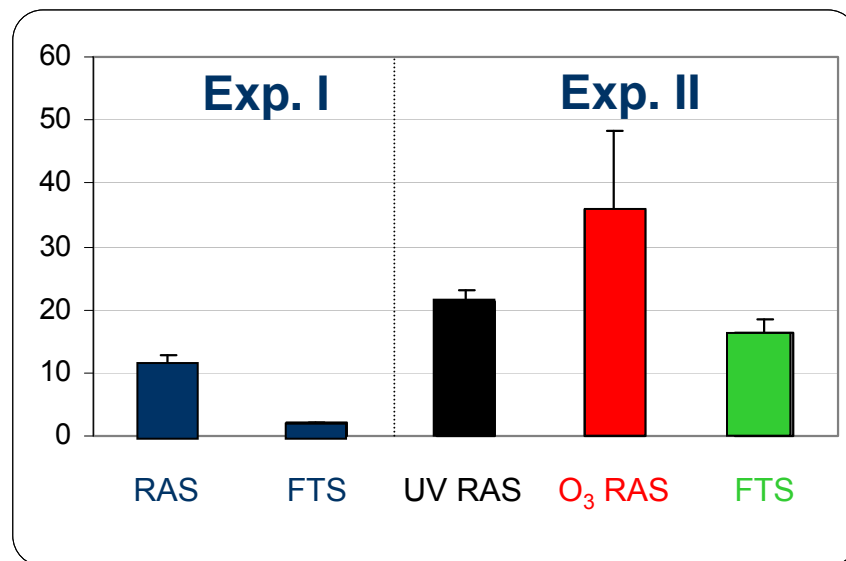


Result 5: Equal or better fish survival in RAS

Density by photography



Survival through live feed period (%)



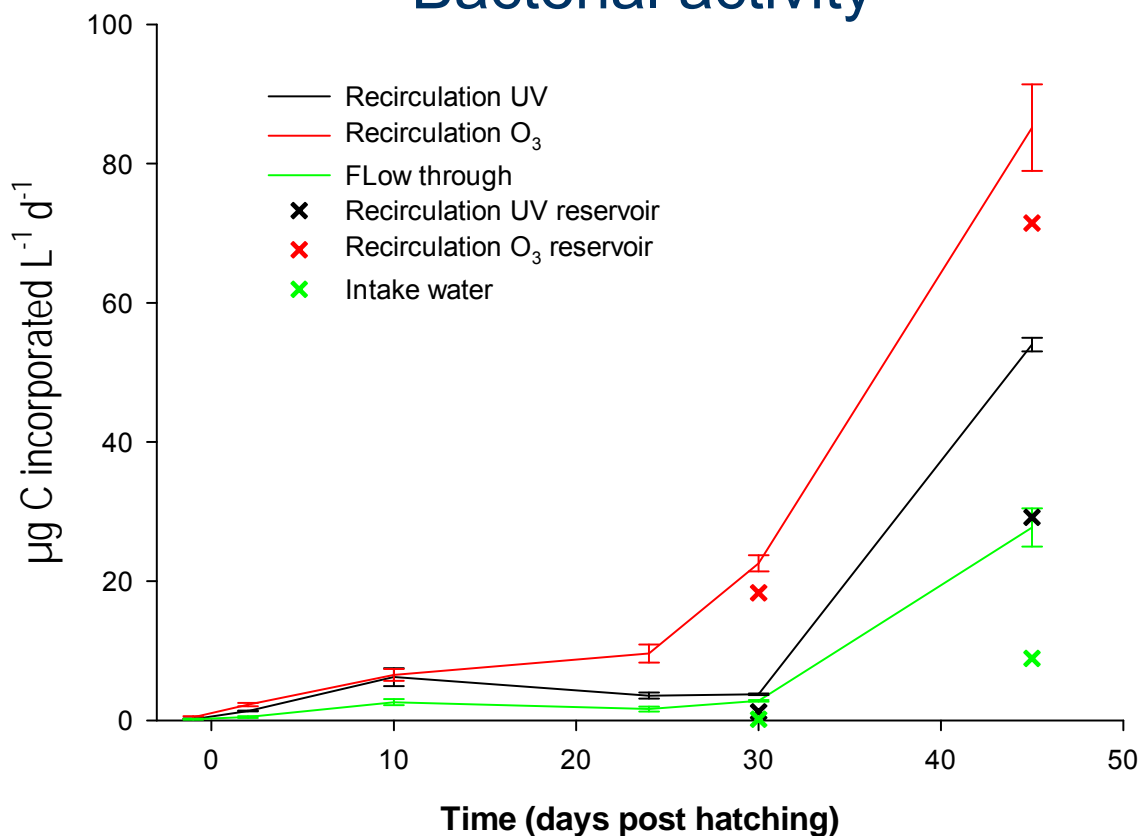


Result 6: Less RAS disinfection - better results

**Microbial activity in tanks
compared to
inflowing water:**

O₃ RAS 1.2 × higher
UV RAS 3.0 × higher
FTS 17.5 × higher

Bacterial activity





Aknowledgement: Norwegian Research Counsel, NTNU, SINTEF