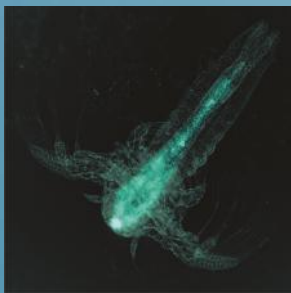
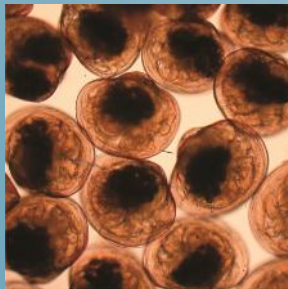
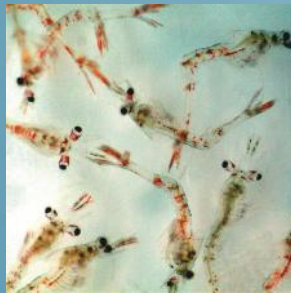
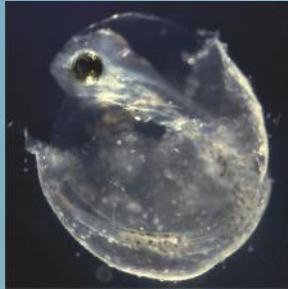


larvi 2013

6th fish & shellfish larviculture symposium



Control of the selective pressure
on microbes in rearing tanks
through manipulation of the incoming water
increases survival of marine fish larvae



Kari Attramadal

ghent university, belgium, 2-5 september 2013

Control of the selective pressure on microbes of the incoming water increases survival of marine fish larvae

Attramadal, K.^a, Øie, G.^b, Kjørsvik, E.^a, Minniti, G.^a, Skjermo, J.^b, Bakke, I.^c, Østensen, M-A.^b, Olsen, Y.^a, Vadstein, O.^c

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^c Department of Biotechnology, NTNU, 7491 Trondheim, Norway



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Marine fish larvae

- Period of rapid growth and development of organs
- The specific immune system not fully functional until after metamorphosis
- Sensitive to infections



Common (microbial) problems in larviculture?



- Often large tank to tank variations in survival despite same egg group, feed and physicochemical conditions
- Antibiotics increase reproducibility of survival
- The rearing water is an important source of bacteria for the larvae



Microbial control of rearing water!



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Microbial control: Specific pathogens and opportunists



- Specific pathogens may be stopped by **strong hygienic barriers** into the system:
BIOSECURITY!



- A lot of the problems in aquaculture caused by naturally occurring opportunistic bacteria that become pathogenic when the host is weakened by environmental stress
- It is possible to set up selection to **outcompete the opportunists!**



Generally one out of two different strategies favoured: Ecological r/K-theory

Carrying capacity (CC)

= Max biomass/number of bacteria that can be maintained in the system over time

Depends on:

Supply of available organic matter

Selection	Environment	Substrate supply per bacteria	Favoured ability
r-selection	Unpredictable/unstable, Empty niches	High	Rapid reproduction, Fast growth → Opportunist
K-selection	Stable or predictable, crowded	Low, Close to CC	Competing on limited resources → Specialist



↑
Colonising bacteria
typically r-strategist

↑
Succession: bacteria with
increasing ability to compete

↑
Opportunists gradually
replaced by K-strategists

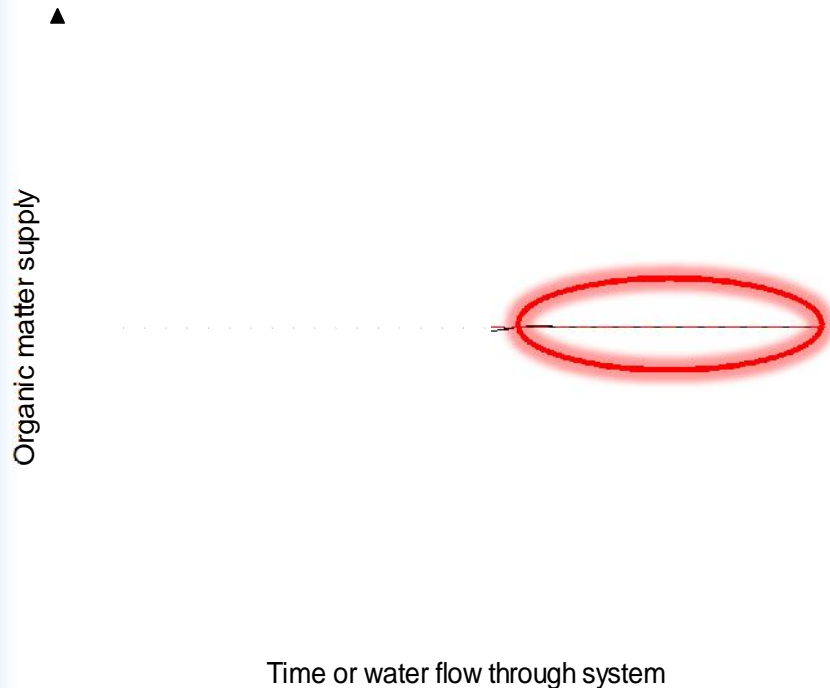


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K-selection: Microbial maturation

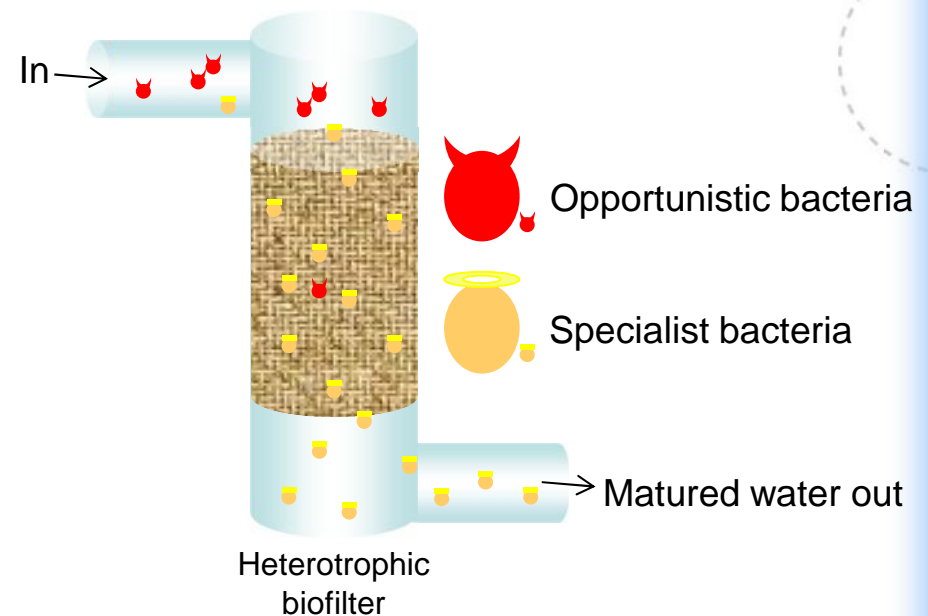
How?

Wait for succession at a given CC



Maturing unit = biofilter

Low substrate supply per bacteria, favouring the specialists over the opportunists



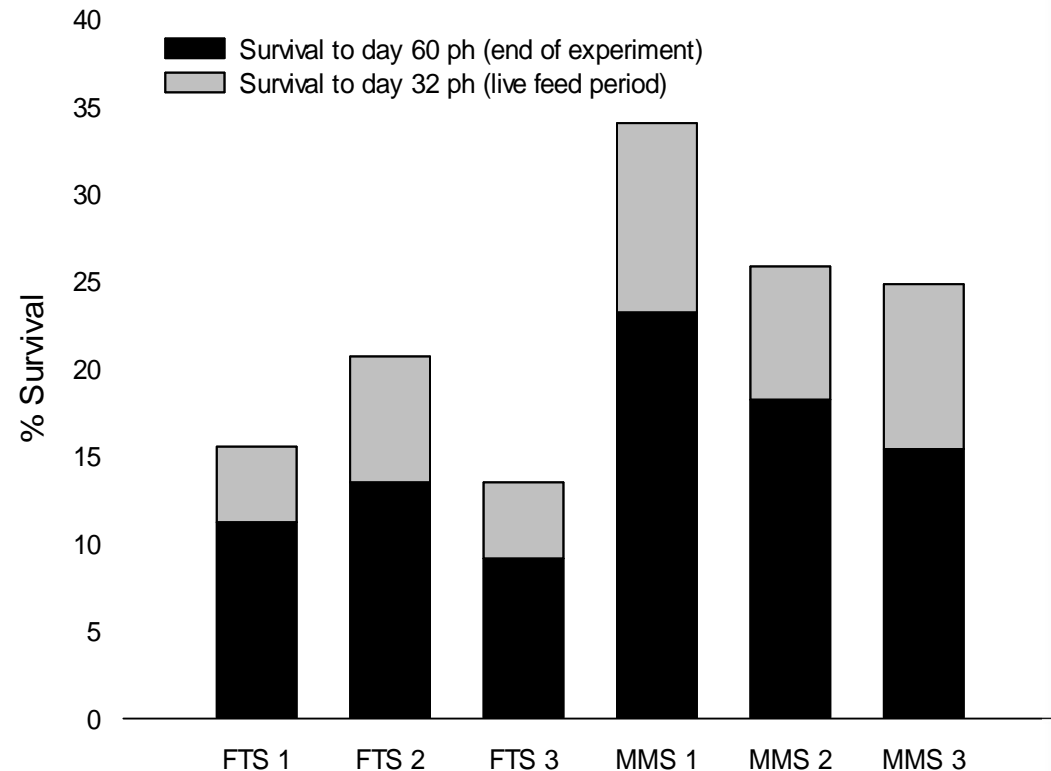
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K-selection: Microbial maturation

Effects on the microbial community composition of the incoming water:

A more stable, even and diverse community dominated by slow-growing specialists

Effect on the fish: Significantly higher survival



Incoming water

Tank water

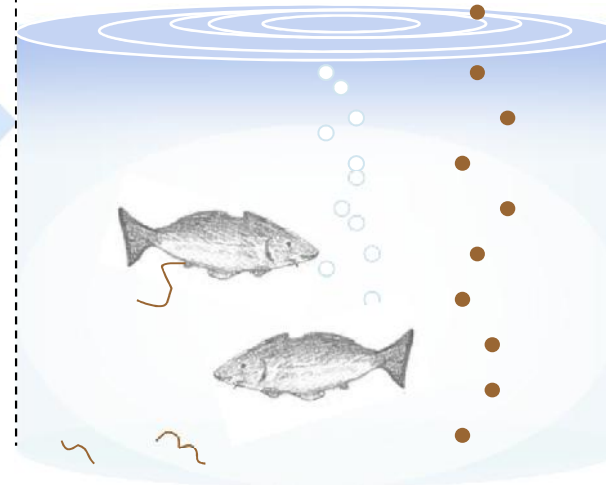
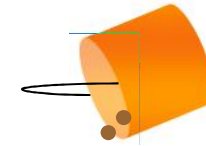
Selection pressure



- Intake bacteria
- Disinfection
- Intake org.matter
- Particle removal

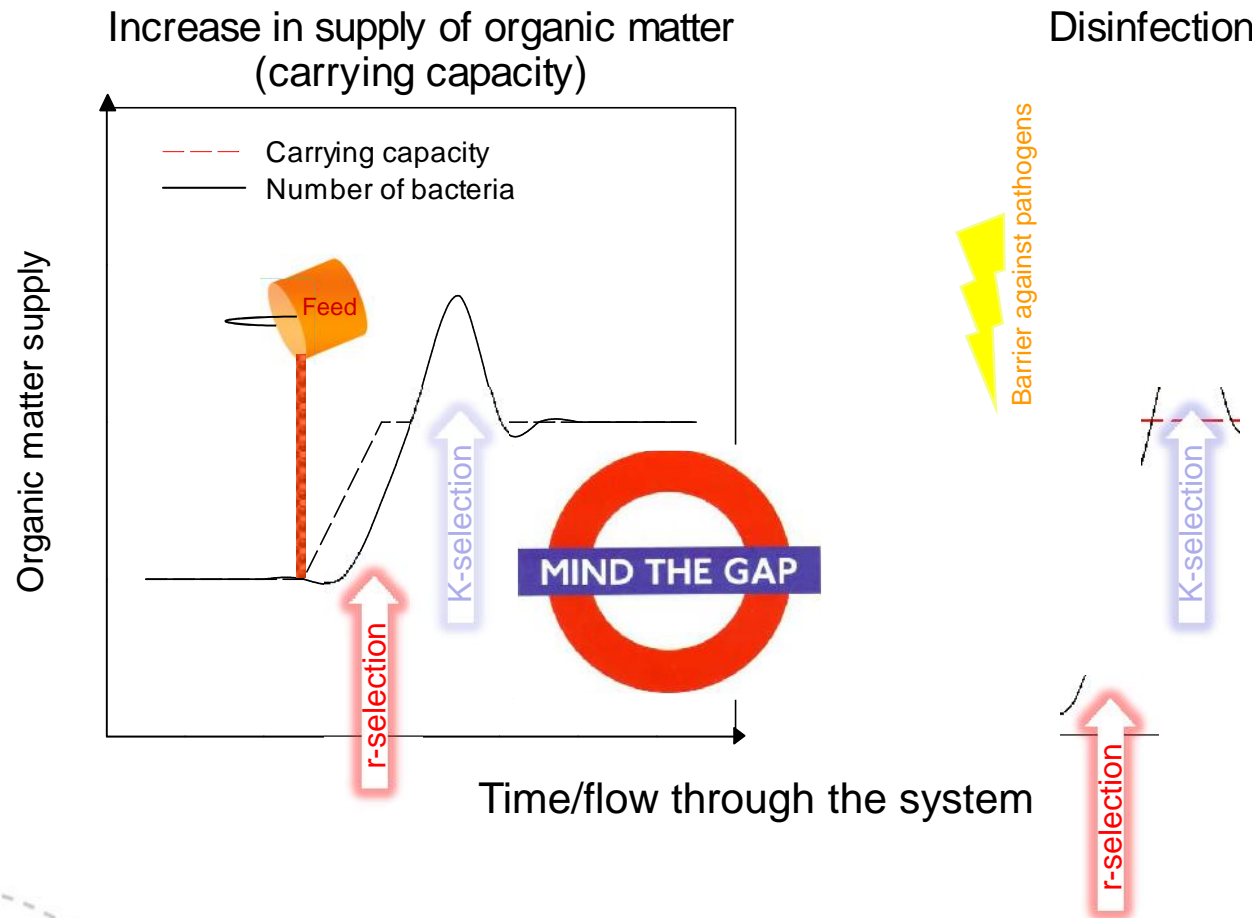


- Water exchange rate
- Feeding
- Faeces



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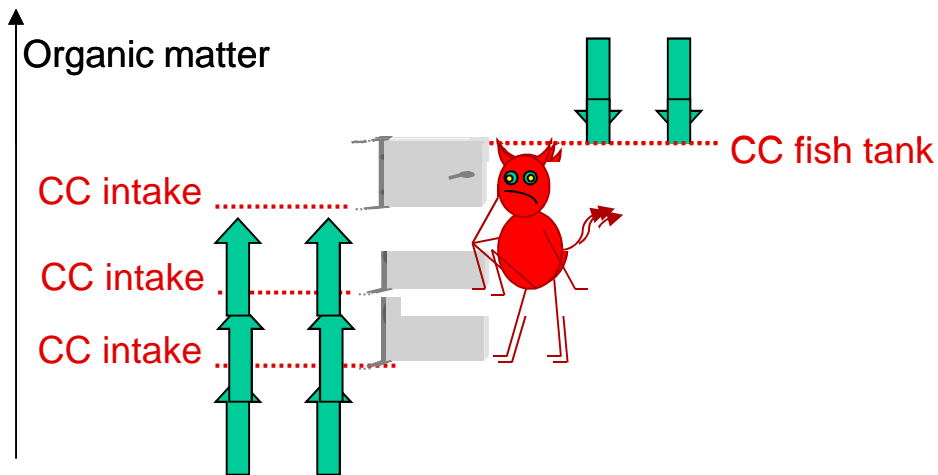
Perturbances promoting r-selection in intensive aquaculture



Closing the gap:

K-selection/maturation should be carried out at a carrying capacity (CC) (organic matter supply) similar to that of the rearing tanks

Increase CC in the matured inflowing water Reduce CC in the rearing tanks



1. Recirculation (RAS)
2. Feed the maturation filter (flow through)

1. High water exchange rates
2. Efficient cleaning
3. Addition of clay instead of algae



Closing the gap - gaining control

K-selection/maturation of the microbial community of incoming water at a carrying capacity (CC) similar to that in the tanks

1. RAS
2. Feed the maturing unit (flow through)

Reduce the use of disinfection inside the RAS

1. In the recycling loop
2. Before tanks



Closing the gap 1

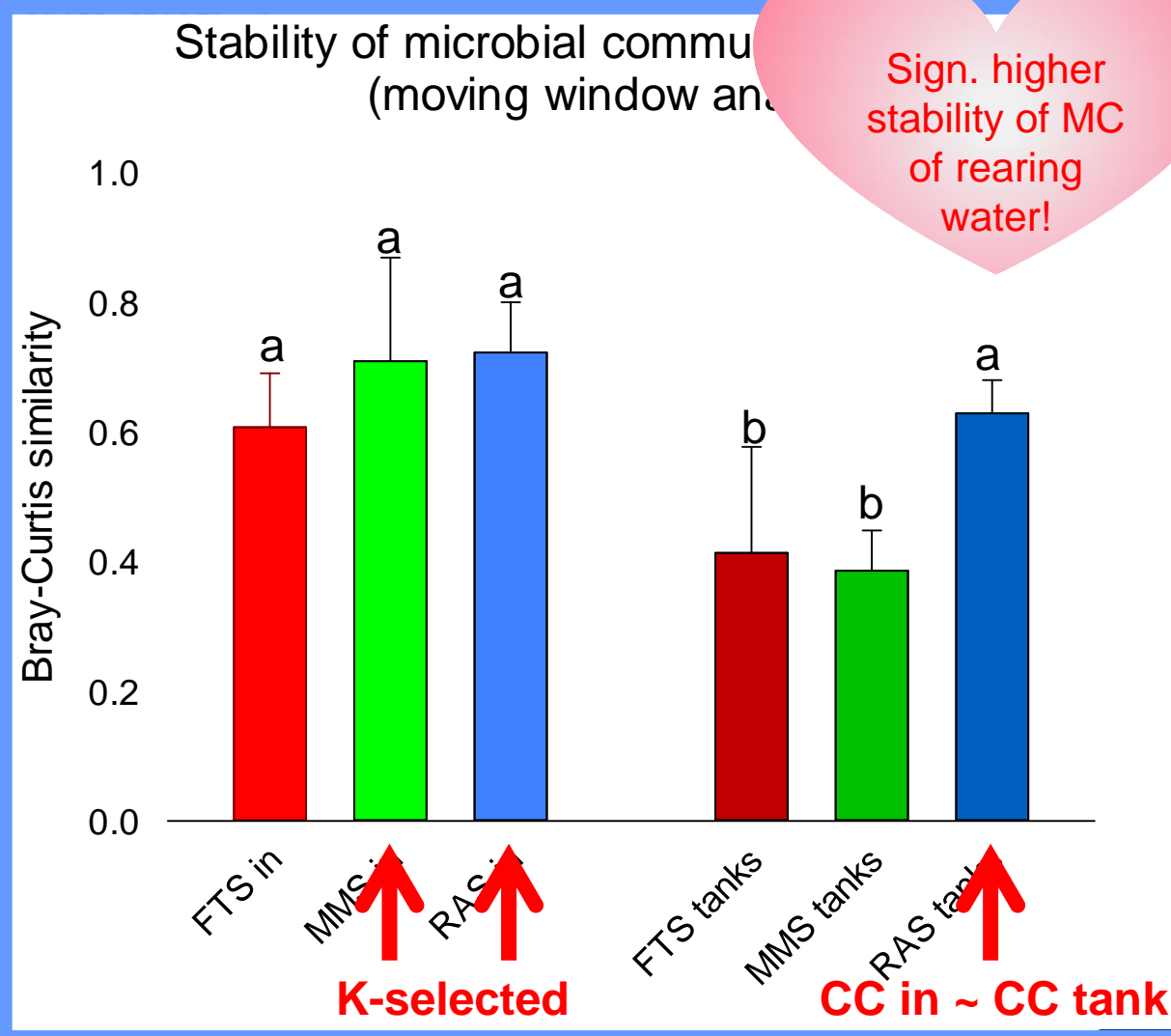
RAS

System
bacteria in
Bacterial bio

FTS: no tanks lea

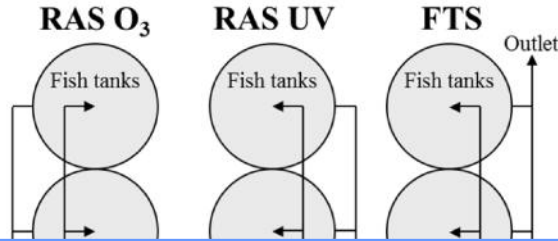
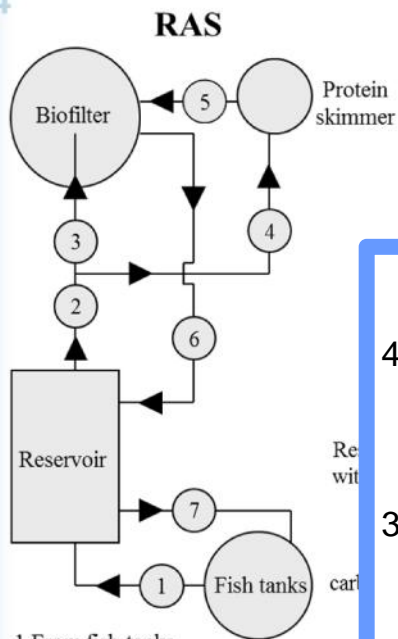
MMS: ma open for

RAS: ma buffer ag

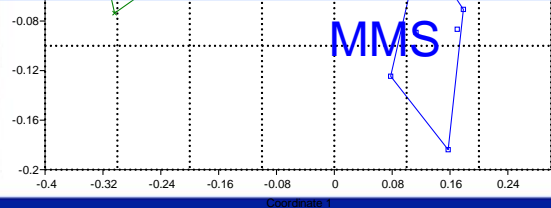
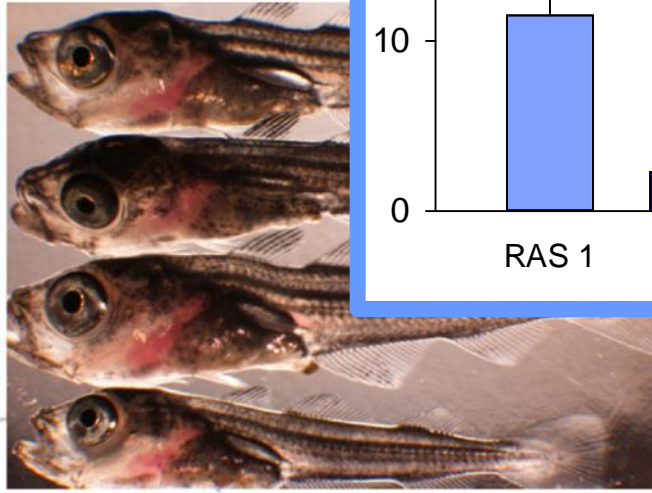
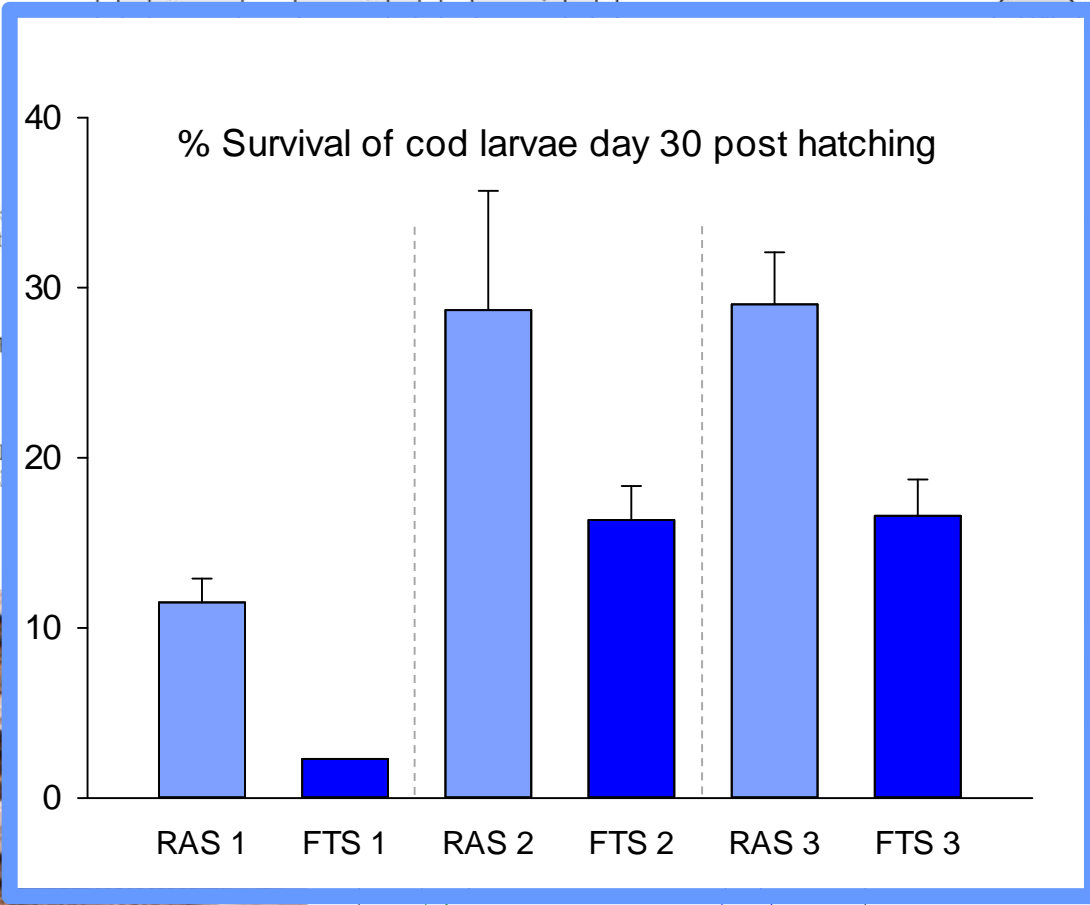
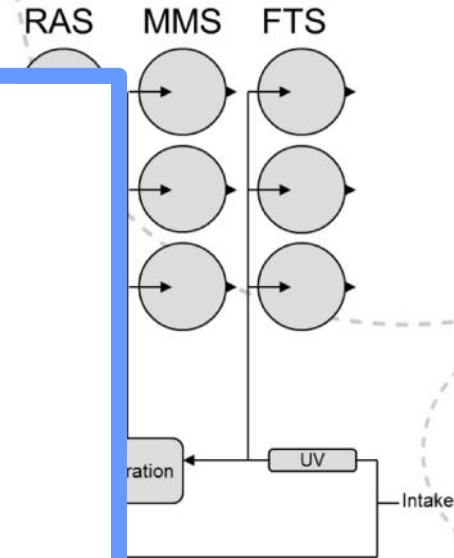


RAS
65 ± 4
29 ± 6

of
may
longer



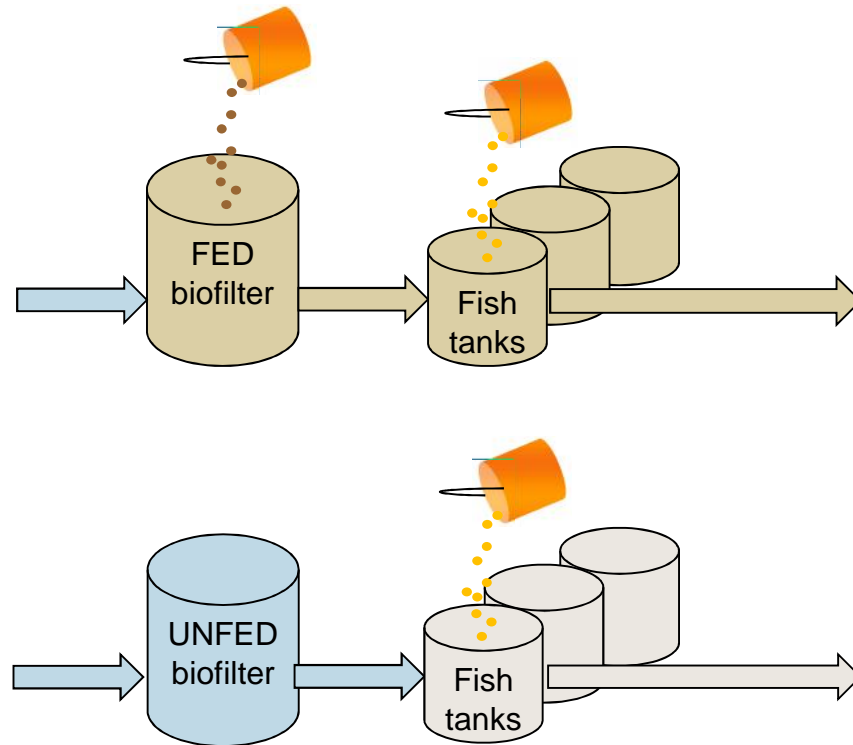
RAS



ter microbial
ity composition
ble, diverse
n in the RAS

Closing the gap 2

Feed the maturation filter (flow through)

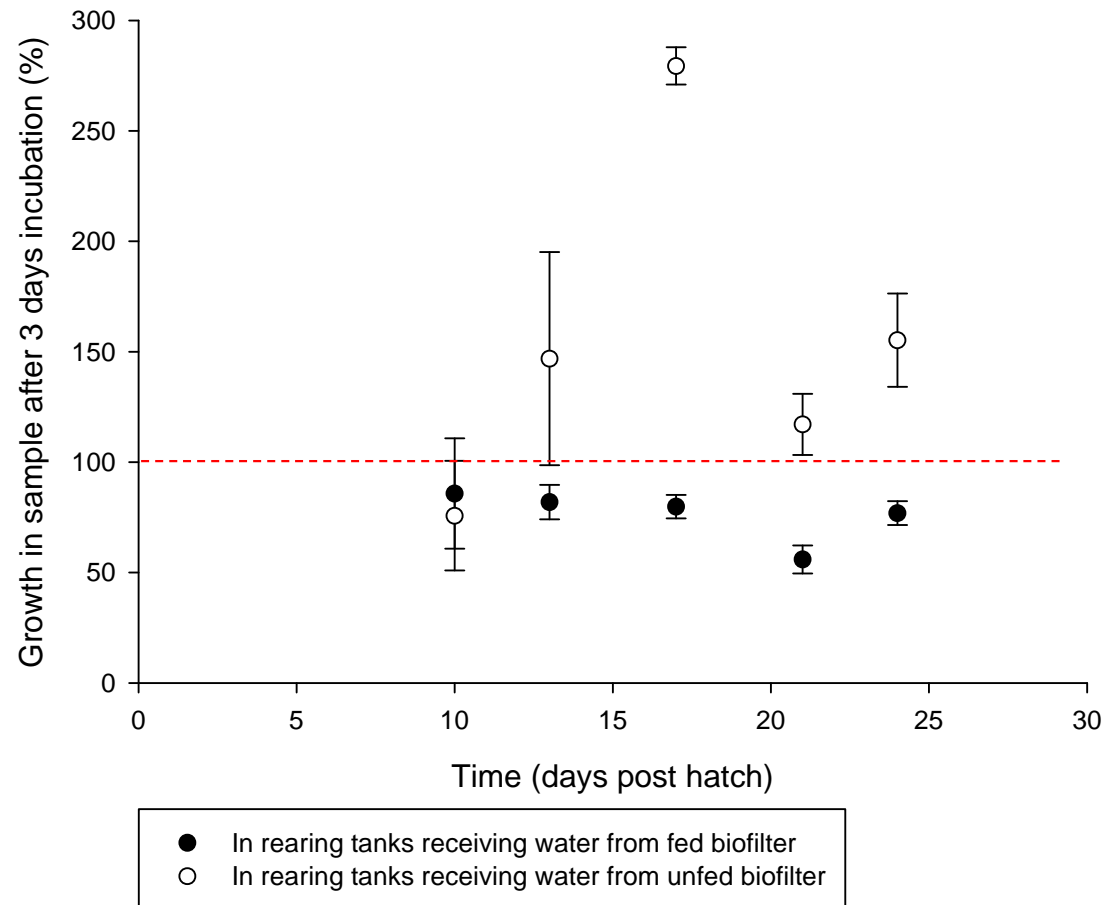


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Closing the gap 2

Feed the maturation filter (flow through)

Microbial growth potential in tank water (3 days)



Increased # bacteria
after 3 days of
incubation – growth
potential in the sample

No net growth

No growth potential:
crowded environment
resistant to invasion



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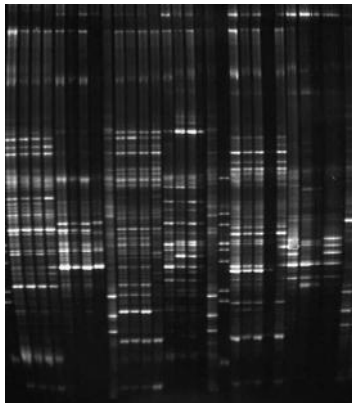
Closing the gap 2

Feed the maturation filter (flow through)

Microbial community composition

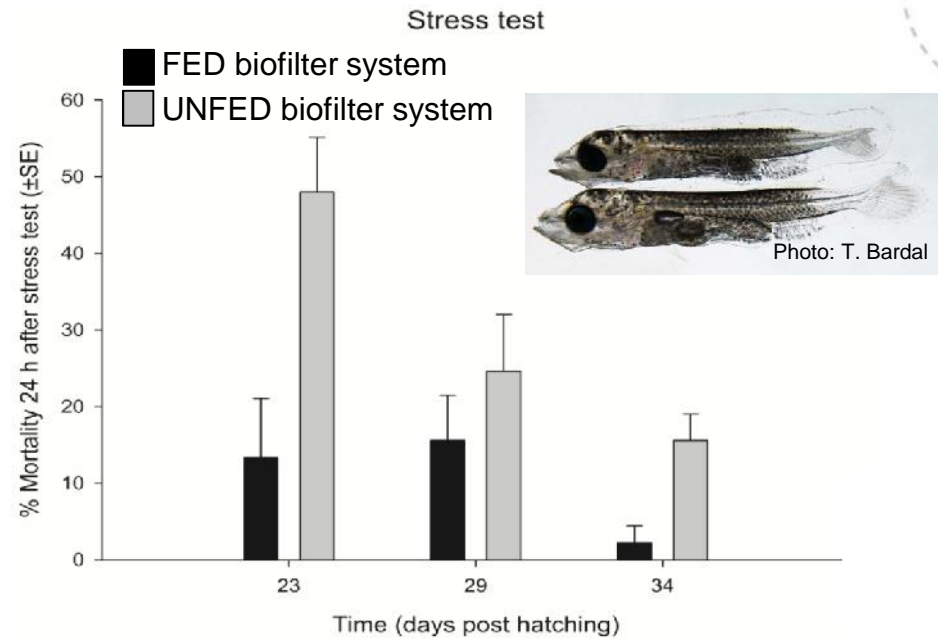
- Tank water sign. more similar to intake water in FED
- Replicate tanks sign. more similar in FED
- Stability in tanks over time, species richness, diversity (Shannon's index H') and evenness (J') significantly higher in tanks in FED

DGGE



Larval performance

- Survival (27 dph): 5% in FED, 20% in UNFED
- Stress tolerance: significantly higher in FED
- Growth: significantly higher in FED*



Reduce the use of disinfection inside the RAS 1

In the recirculation loop

Strong disinfection within the RAS loop may reduce the maturing effect of the microbial community:

FTS: high disinfection efficiency on the incoming water lead to to proliferation of bacteria in tank

RAS UV: high disinfection efficiency reduce abundance and activity of bacteria in incoming water and may open for proliferation in tank

RAS O₃: ozonation to 350 mV in protein skimmer results in low or no disinfection and an incoming water similar to the water in tanks

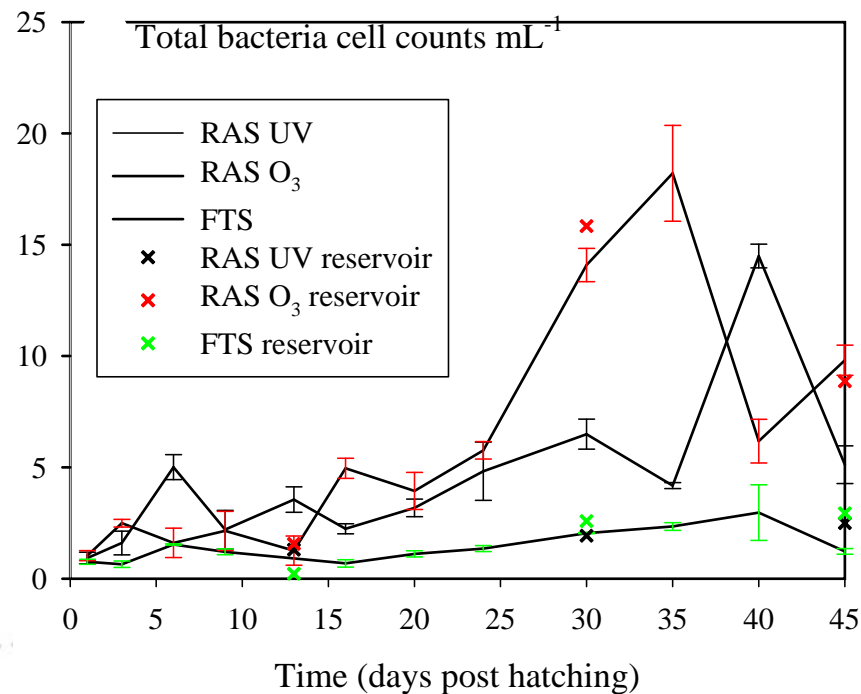


Reduce the use of disinfection inside the RAS 1

In the recirculation loop

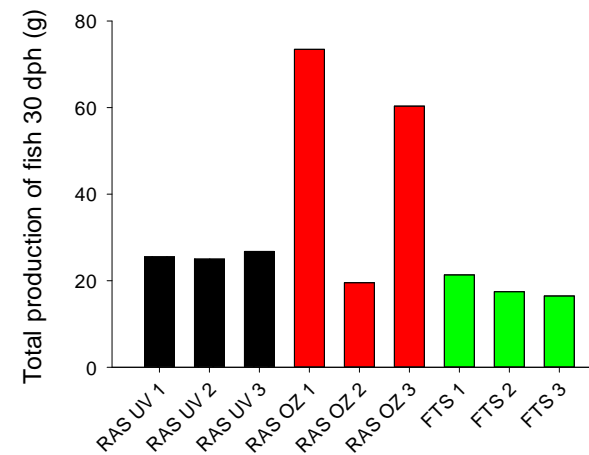
RAS_{O₃} more mature and stable microbial community than RAS_{UV}

Incoming water microbial community composition more similar to tank water in RAS_{O₃} compared to RAS_{UV} and FTS:



Microbial activity (production of bacterial biomass) in tanks compared to inflowing water:

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

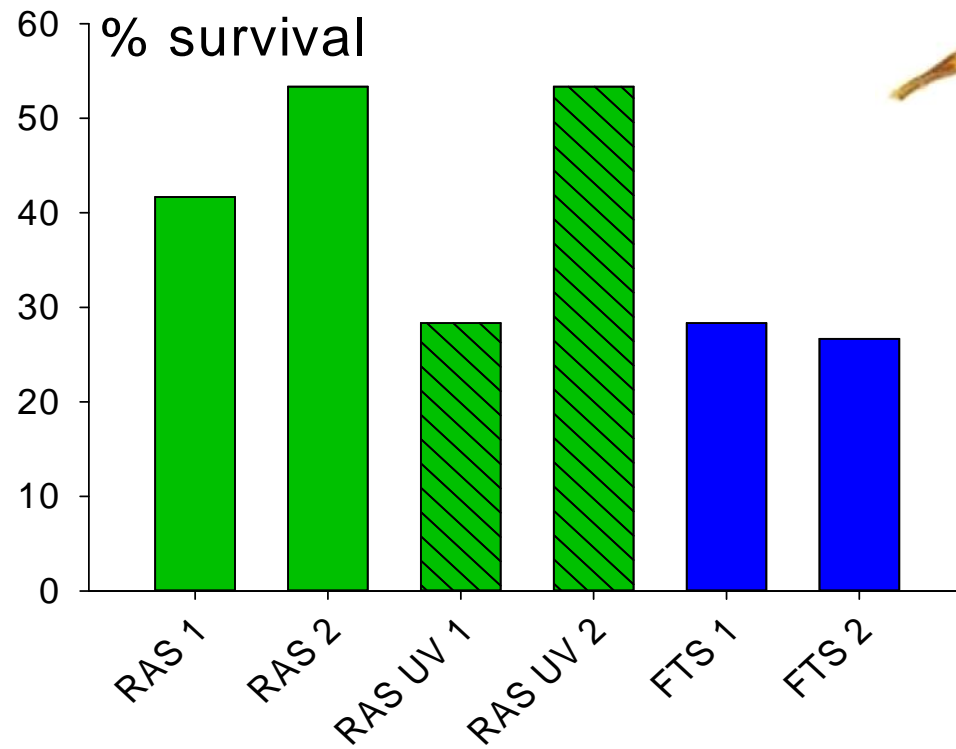


Reduce the use of disinfection inside the RAS 2

Before rearing tanks

(From John Vegard Øien)

Survival of lobster 14 dph



Best survival in RAS with no disinfection

Microbiota yet to be analysed



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Conclusions

Similarity of the selective conditions for the microbes in the incoming water and in the rearing water is a key to microbial control in the fish tanks, and opens for optimization of water treatment of the incoming water to benefit larviculture

Controlling the microbial carrying capacity in the different components of the system is a very good idea!

Using strong disinfection on the incoming water can be smart (biosecurity), but avoid it in the recirculation loop!



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Recycle!

Thank you for listening!

Kari Attramadal



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