

# Magic solutions to bacterial problems in early life stages: Do they exist?

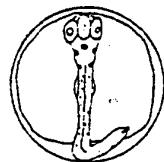
Ingrid Salvesen, Jorunn Skjermo and Olav Vadstein

SINTEF Fisheries and Aquaculture, Norway

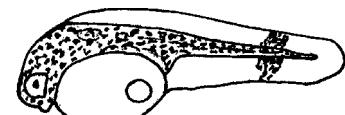
NTNU, Trondheim Biological Station, Norway

# MICROBIAL CONDITIONS

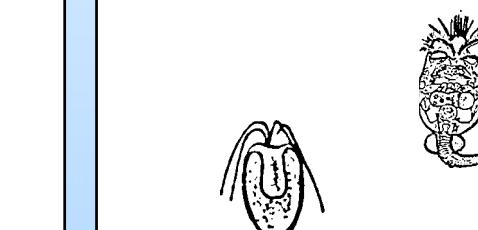
EGG



YOLK SAC



FIRST FEEDING



two-step

three-step

Physical, chemical factors: temp., exchange rates, water quality...

# MICROBIAL CONTROL??

- It is a **dynamic** system!
- High reproductive rates and metabolic versality - high speed of adaptive changes
- Short generation times - large population fluctuations
- Capacity for dormancy
- Beneficial mutations can be rapidly established by natural selection
- Various strategies for survival:
  - Not resource limited - high growth rates
  - Resource limited - many competitive adaptations



# MICROBIAL CONDITIONS



## Early life stages

- Sterile at hatching - the importance of the bacterial load and the composition of the ambient water for the establishment of a primary protective microflora
- Bacteria and microalgae ingested at the yolk sac stage
- Differences between marine fish species (short vs long yolk sac period, clear vs green water)
- Exposure to stressors in early life stages - adverse effects later in development



# HATCHING

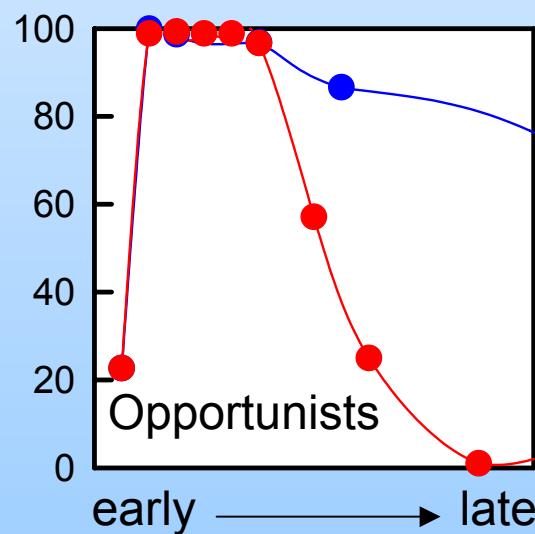
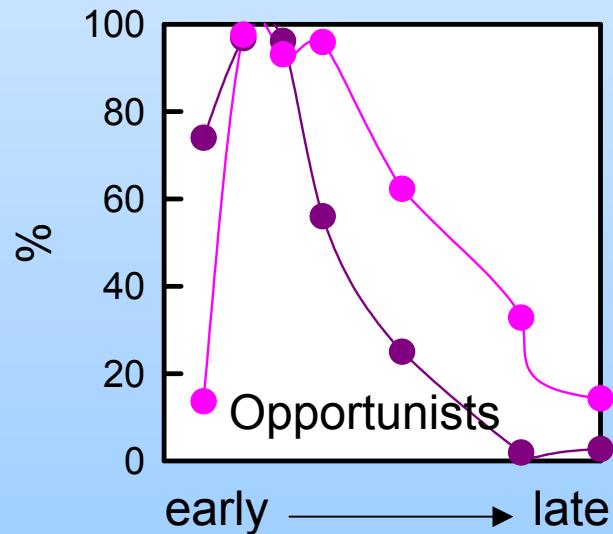
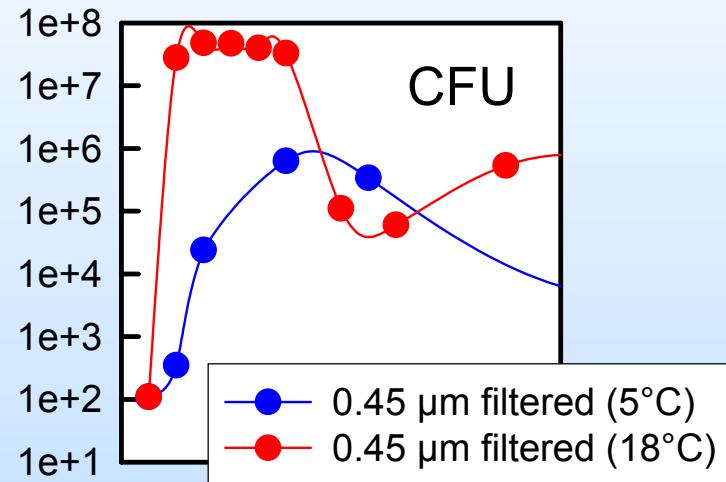
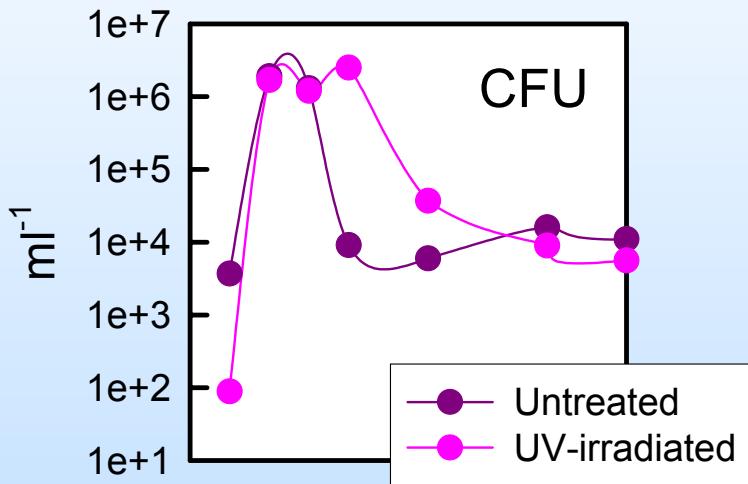
## Enhanced organic loading

### CARBON BUDGET

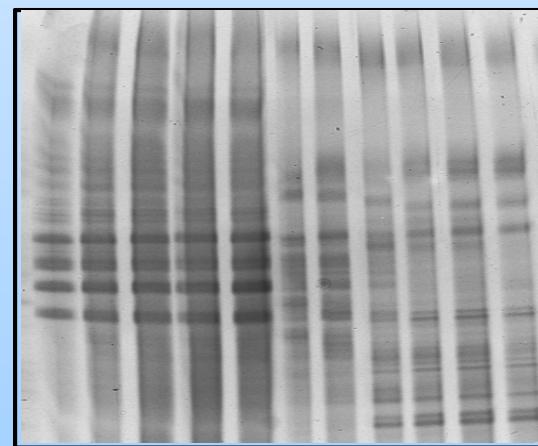
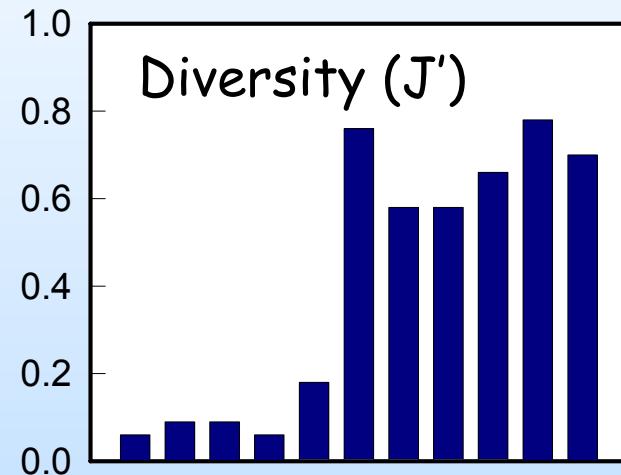
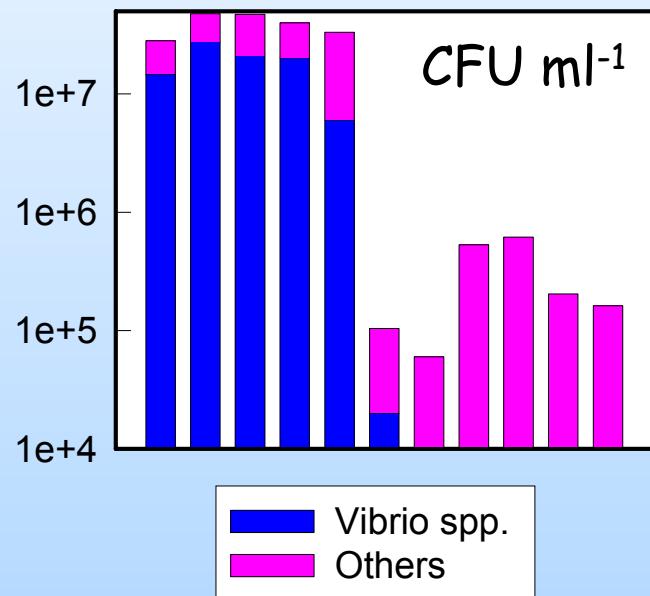
	$\mu\text{g C egg}^{-1}$	%
Larvae	552	65.0
Chorion	12	1.4
Estimated DOC loss	286	33.7

Dahlø et al., unpublished results

# No bacteria introduced, only substrate...



# No bacteria introduced, only substrate...



Beat them  
or  
join them?

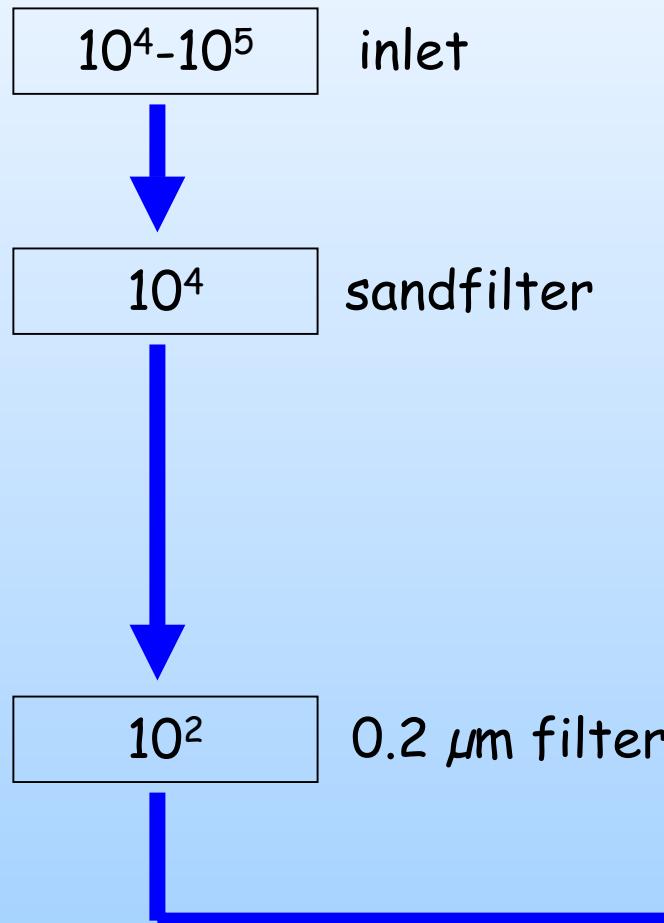
*The Unknown Enemy*



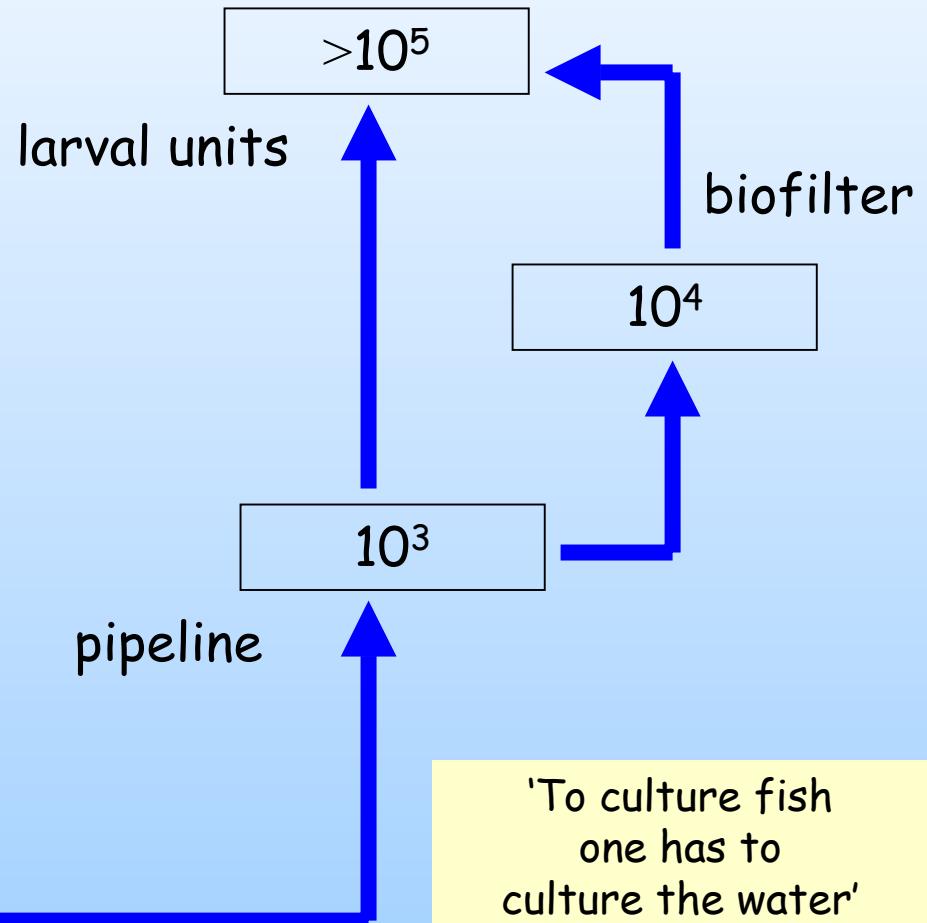
*Habit des Medecins, et autres personnes  
qui visitent les Pestiferés. Il est de  
marroquin de leuant, le masque a les yeux  
de cristal, et un long nez rempli de parfumis.*

*Fig. 2. Protective clothing of the kind worn by  
plague doctors during the seventeenth century.*

# REDUCTION



# RECOLONIZATION



# RECOLONIZATION

## A probiotic approach?

MICROALGAE - green water technique

# ALGAE

## Interactions with bacteria



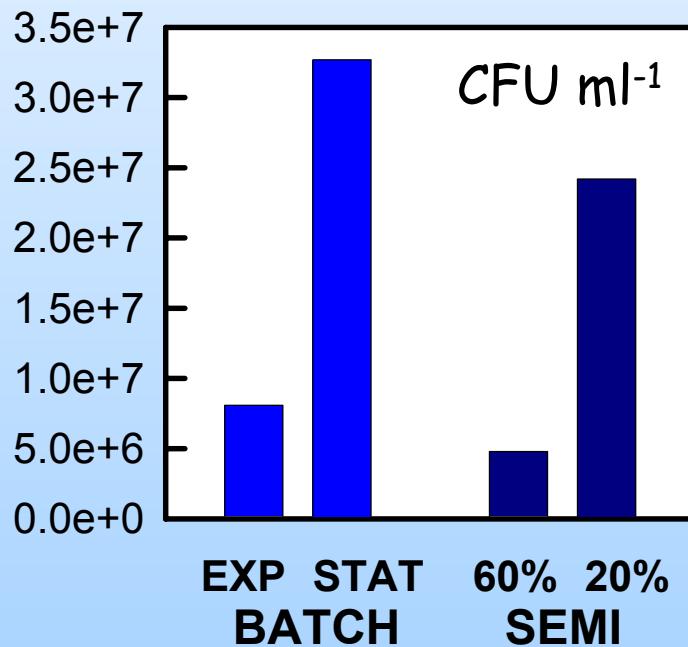
- stimulate growth by release of dissolved organic carbon
- inhibit growth through production of toxic metabolites
- interfere with the expression of traits important for virulence, colonization

A wide range of mechanisms by which algae may regulate bacterial communities

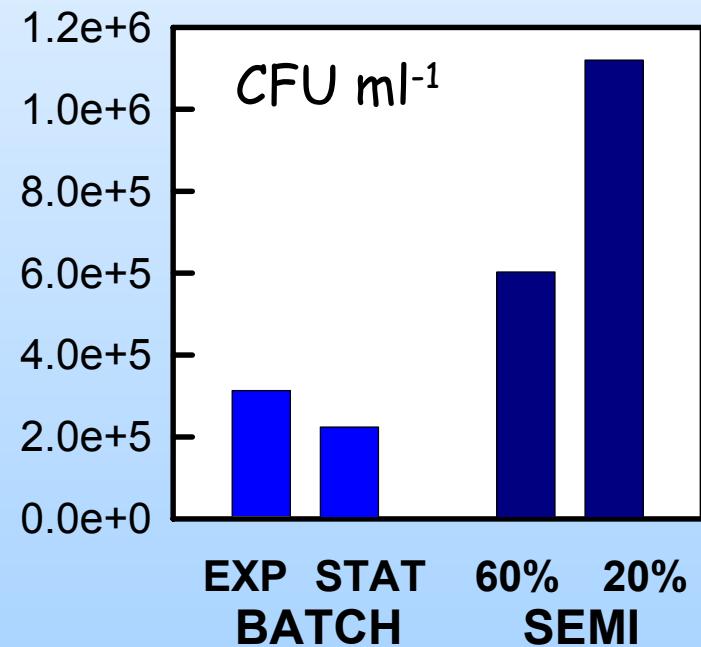
# BACTERIAL LOAD

## *Isochrysis galbana*

Associated with algal culture



Added to obtain a algal density of 100 cells  $\mu\text{l}^{-1}$  in larval culture



No (batch) or very low levels (< 0.01%, semi-continuous) of *Vibrio* spp.

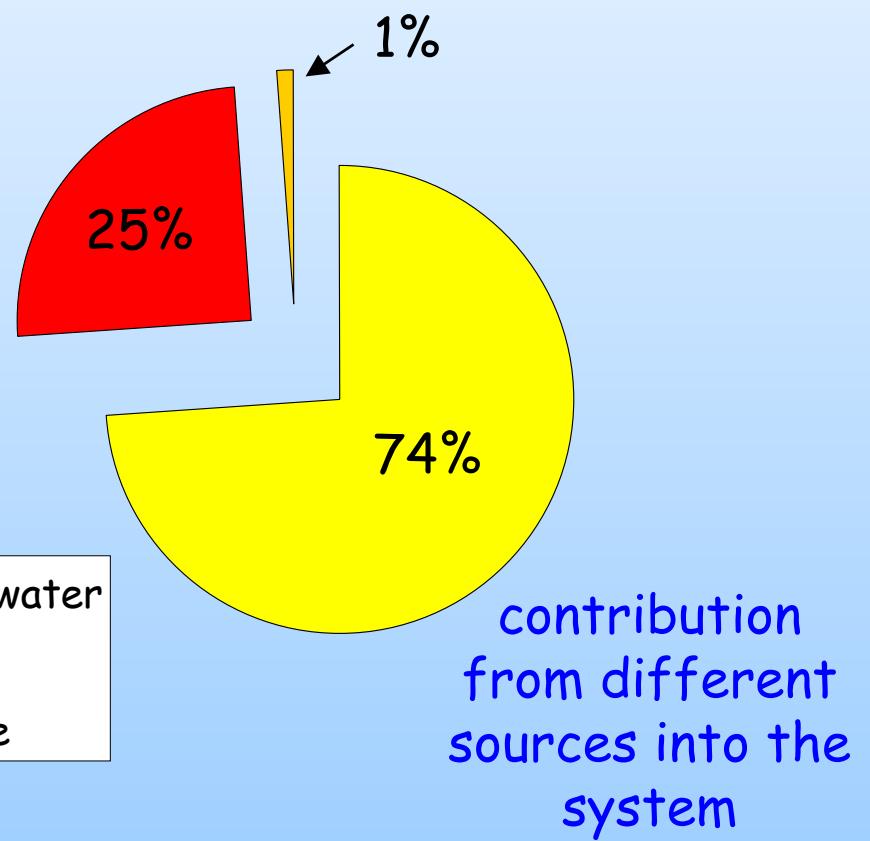
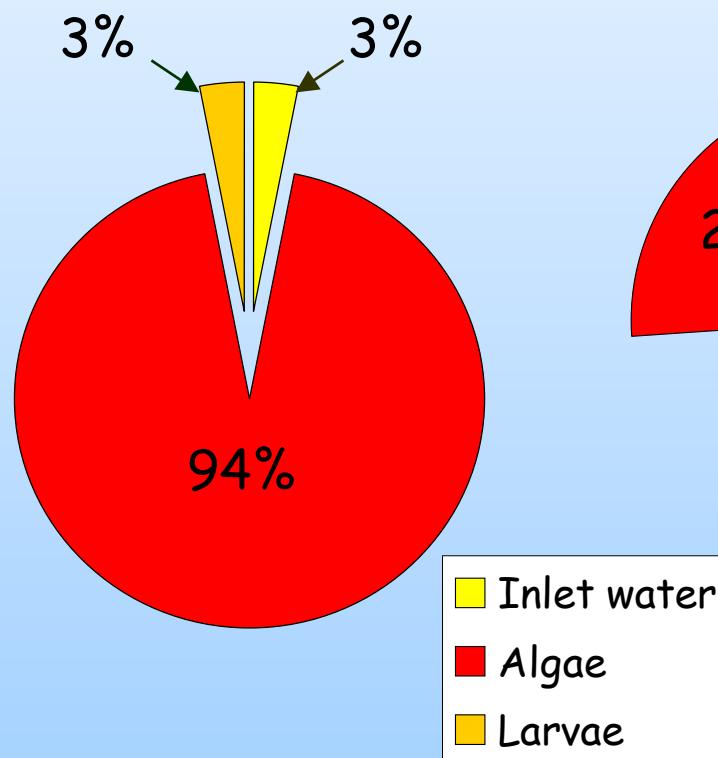
# Contribution of bacteria

bacterial level

in inlet water:

$1 \times 10^3$

$1 \times 10^5$

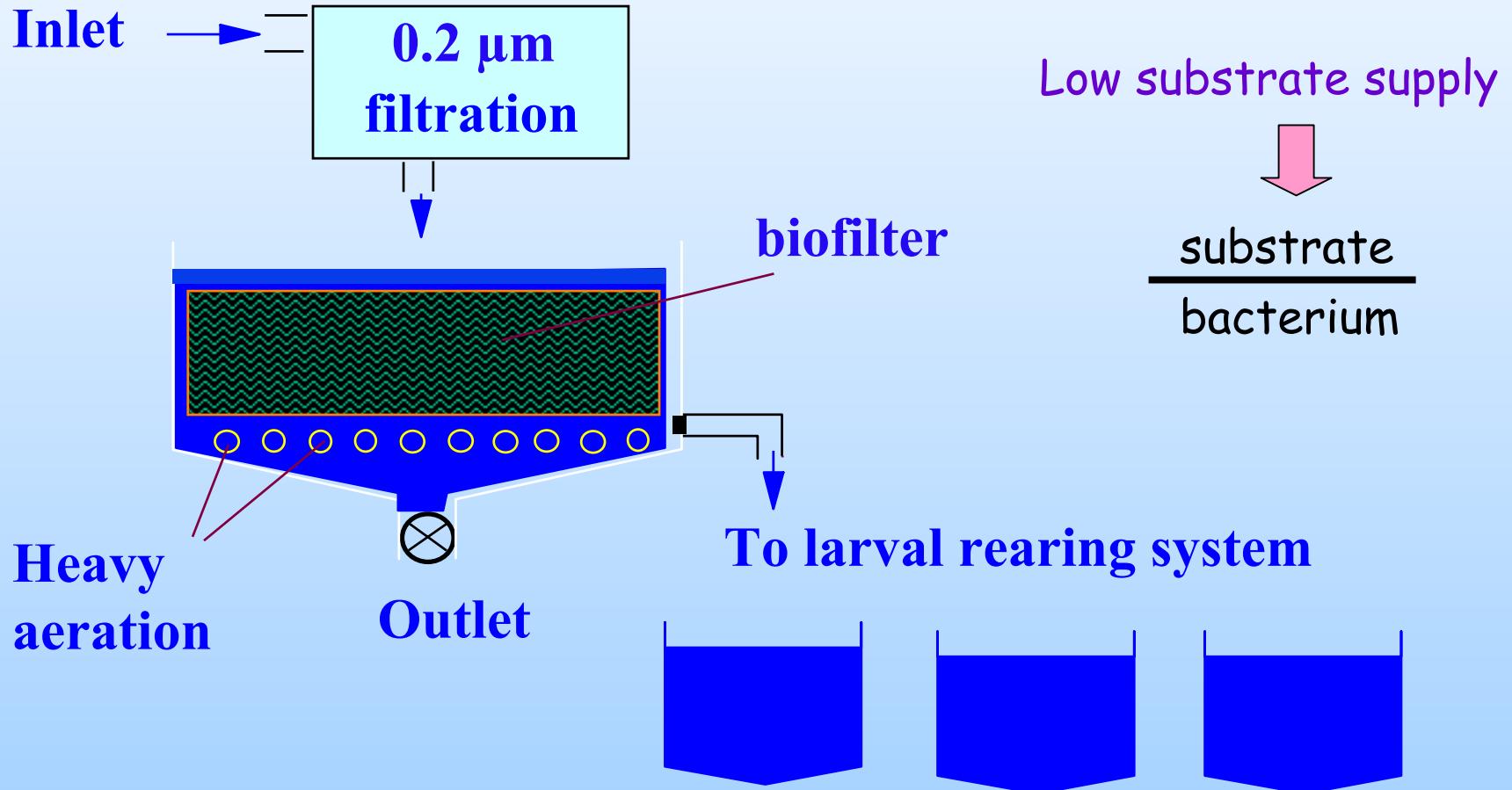


# RECOLONIZATION

## A probiotic approach

WATER TREATMENT - biofilter technology

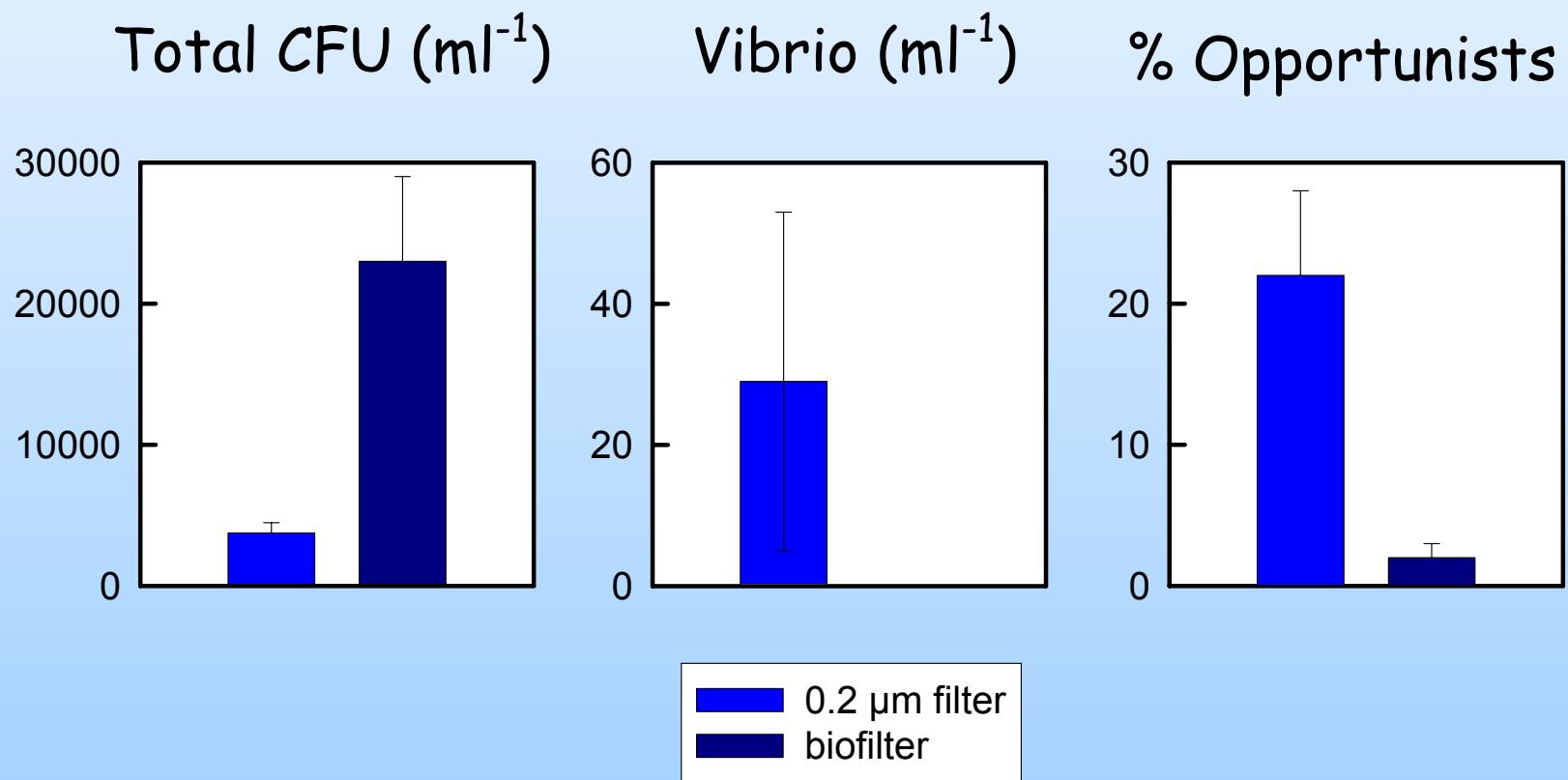
# BIOFILTER (flow-through)



Skjermo et al., 1997

# RECOLONIZATION IN BIOFILTER

## A magic solution?



# TWO WATER QUALITIES

## 0.2 $\mu\text{m}$ filter and biofilter

How is the bacterial community of the water  
affected by enhanced organic loading in  
combination with microalgal addition?

# ORGANIC LOADING

	Total CFU		Vibrio		% Opport.	
	0.2 µm	biofilter	0.2 µm	biofilter	0.2 µm	biofilter
Clear water	$2.1 \cdot 10^7$	$1.3 \cdot 10^6$	$4.0 \cdot 10^6$	$9.5 \cdot 10^3$	100	99

Low levels of opportunistic bacteria (0.3-7%) and no Vibrio spp. observed in both water qualities before organic loading

Everything is everywhere,  
the environment selects

M.W. Beijerinck

# ORGANIC LOADING

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Clear water	$2.1 \cdot 10^7$	$1.3 \cdot 10^6$	$4.0 \cdot 10^6$	$9.5 \cdot 10^3$	100	99
Green water						
<i>Isochrysis</i>	$2.5 \cdot 10^7$	$9.8 \cdot 10^5$	$5.4 \cdot 10^4$	$1.2 \cdot 10^3$	98	69
<i>Pavlova</i>	$2.0 \cdot 10^7$	$5.4 \cdot 10^6$	$2.0 \cdot 10^6$	$3.0 \cdot 10^2$	88	8

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<i>Chaetoceros</i>	$1.5 \cdot 10^7$	$2.4 \cdot 10^7$	$1.3 \cdot 10^6$	$1.9 \cdot 10^5$	60	19
Mix (1:1:1)	$1.4 \cdot 10^7$	$1.3 \cdot 10^7$	$8.2 \cdot 10^5$	$5.9 \cdot 10^5$	83	44

Relatively high bacterial densities and large proportions of opportunistic bacteria are associated with cultures of Bacillariophyceae (e.g. *Chaetoceros* sp.)

# CONCLUSIONS

- The significance of microbial conditions in early life stages
  - exploit growth potential
- Important differences in response to enhanced organic loading
- No magic solutions, but large potentials for microbial management
  - What is removed, will be replaced.....