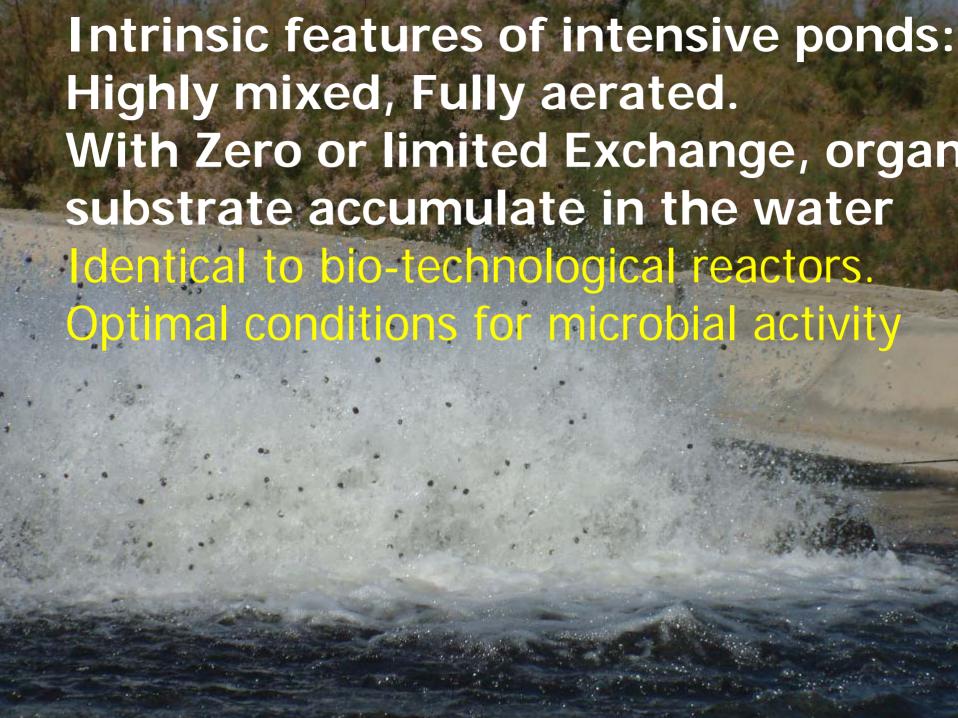


- 1. Limited exchange ponds and build up Of microbial community.
- 2. Control of ammonia
- 3. Bio flocs. Feeding on bio flocs



Characteristics of water:

- 1. Generous supply of feed to microb
- A. Assume 3 kg fish/m3, 15 g feed/day
- ~ 7.5 g C
- B. $\sim 50\%$ released to the water ~ 3.8 g/m3* day
- C. On a steady state basis, with $\epsilon = 50\%$, daily added microbial biomass carbon = 1.9g
- Average cell volume = 0.7 μm3
- Average carbon/cell = 5.6×10^{-16} g
- Computed number of bacteria produced ber day = 4.8 x 10 ⁹/ml

Characteristics of water: 2

- D. Turbid water
- E. Organic carbon in water in the order of 100 mg/l
- F. Number of bacteria counts in ponds
- Around 10⁷ 10⁹ /ml
 - (Compare with computed production of 10^{10} /ml, We see high turn over rate and young microbial population)

Characteristics of water:

 Rate of organic matter degradation in ponds and tanks ~ 0.15 /day (Avnimelech et al., in tanks and 113 commercial ponds samplings) to 0.27/day in laboratory experiments (Torres Beristain 2005), as compared to 0.1-0.2 in waste water treatment plants.

pi.e: 10-20% of the organic matter degrade daily.



The nitrogen syndrome

- Fish use just about 25% of feed nitrogen. The rest excreted. 4 kg of feed protein are needed to produce 1 kg fish protein!!
- Excretion and microbial mineralization generate ammonium. Ammonia is highly toxic.
- Nitrite is also toxic, especially in fresh water systems.
- Ponds are enriched in N as compared to C. Carbon is emitted as CO2. Nitrogen is left in the pond. Can we revert this feature??

- 1. Elevated inorganic N levels in the water is often the limiting factor toward high performance of intensive systems. Inorganic nitrogen concentrations have to be controlled.
- 2. Low efficiency of protein utilization is a waste of money (Protein is the most expensive feed component). In addition, since major source of protein is fish meal, harvested from over exploited oceans, This is also a major

Microbial "nutrition" *Bacteria are made of protein Assume feeding with sugar: CHOH --> CO2 + H2O + energy (~50%) The rest --> Microbial growth Yet, for growth they need nitrogen that is taken from the water!!

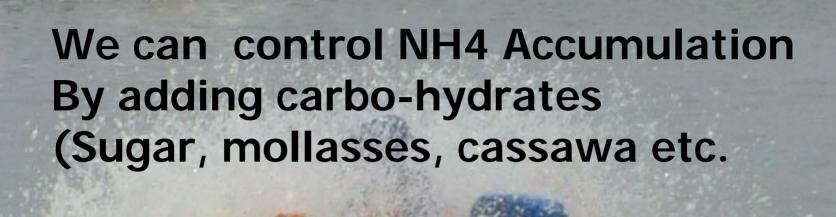
My Diet is Eating
Just Sugar so I stay
slim

I Got fat, I eat sugar + NH4 from the water.











Manipulating bacteria

 We can add carbon rich and protein poor material (carbohydrate, CH), such as starch or cellulose (ground grains, molasses, cassawa etc.) To induce accelerated nitrogen uptake.

Yoram Avnimelech

 $\Delta C = CO2 + \Delta Ccell$ $\Delta Ccell + NH4 ->$ Microbial protein



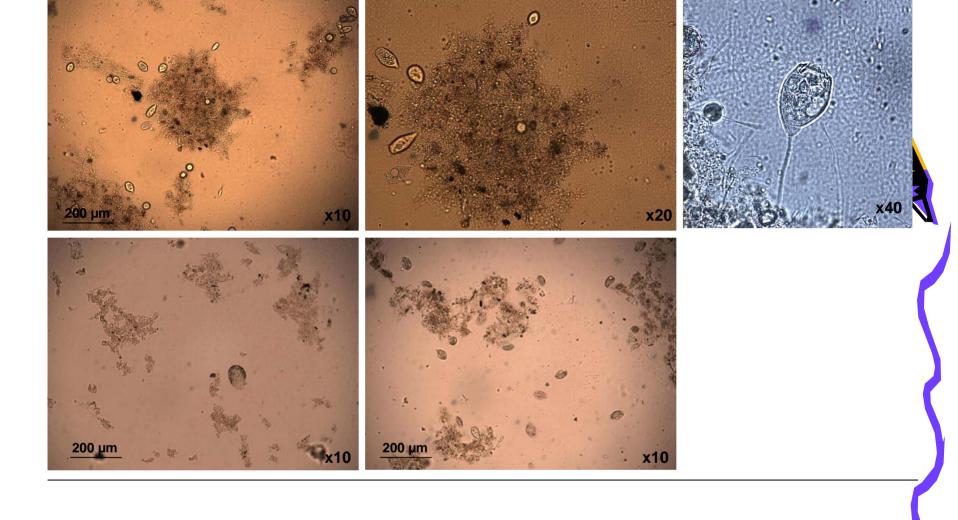
Inorganic nitrogen control is achievable and predictable

Feeding fish with bacteria

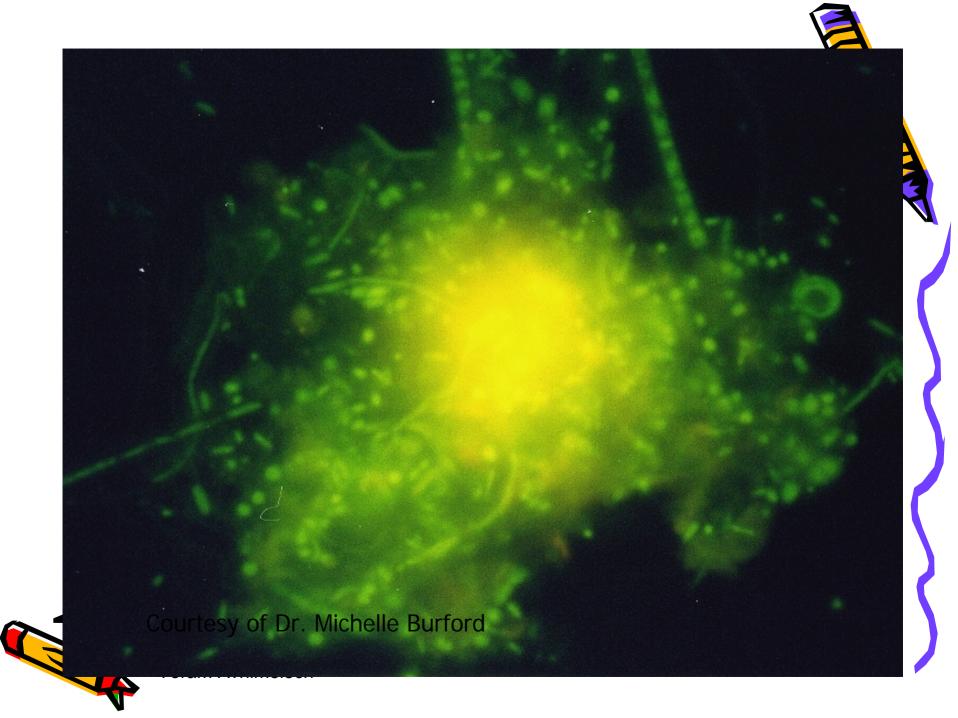
- We can induce the production of microbial protein. Will it be a good feed source for fish???
- Can they physically harvest bacteria? Individual bacteria are too small (~1μm)
- Is it nutritive? Bacterial proteins are different.

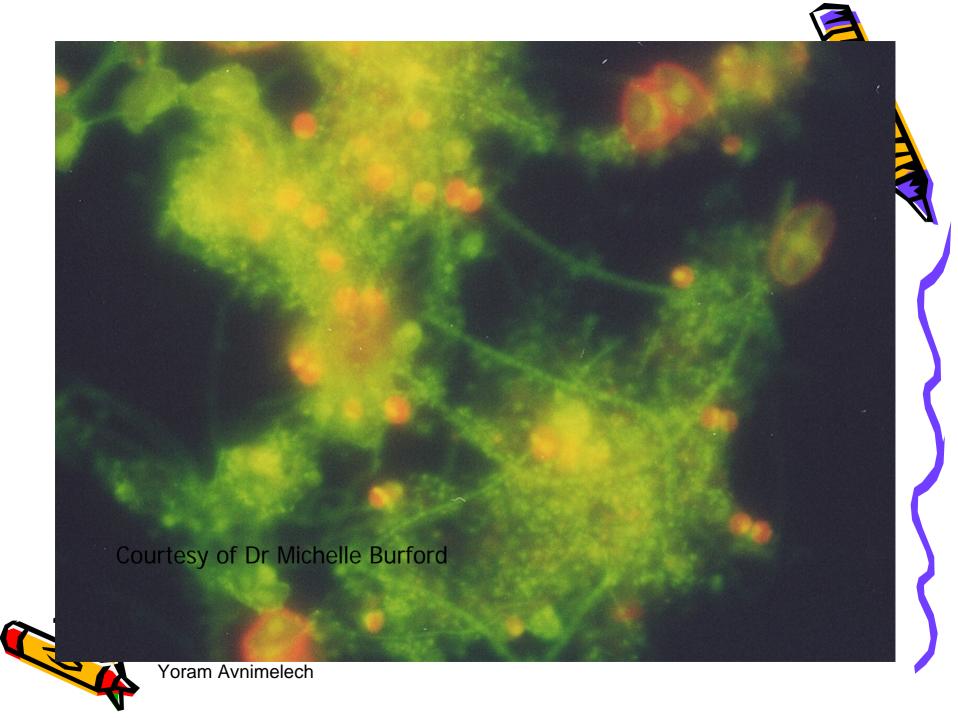
Will they digest it? Probably so





Bio flocs are made of bacteria, protozoa, etc. Typicaly the diameter is 0.1-2 mm.







Results of a study in Belize (Burford et al

The proportion of daily nitrogen requirement of shrimp contributed by natural biota (present mostly as bio flocs) was found, using 15N **take**

study to be

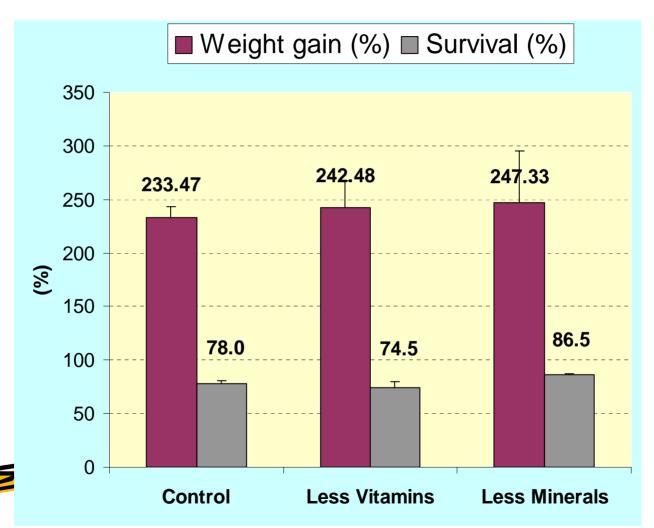


Protein Recycling

- Normally, fish or shrimp recover just ~25% of feed protein.
- In bacterial controlled ponds, they eat the protein twice; Once in the feed and then they consume microbial protein. The protein recovery in experimental and commercial systems reaches almost 50%. An efficiency of 63% was achieved in experimental tanks (Velasco et al.,

Protein is the most expensive part of the feed!!

Results of Albert Tacon (2002)





| Expt. # 1 51 days | 30% Protein | 20% Protein |
|--------------------------|-------------------|----------------|
| FEED C/N | 11.1 | 10. |
| Daily Gain (%) | 1.59 ^a | 2.00 |
| FCR | 2.62 | 2.17 |
| PCR | 4.38 | 2.42 |
| (Kg fish/US\$)FEED COST | 0.848 | 0.583 |
| Exp. # 2 (30 days) | | |
| <u>C/N</u> | 11.1 | 16.6 |
| Daily gain (%) | 1.63a | 2.22^{b} |
| FCR | 2.62 | 2.02 |
| PCR | 4.35 | 2.18 |
| Feed cost (US\$/Kg fish) | 0.848 | 0.543 |

Pros & Cons of microbial N recycling

- 1. Effective, reliable and predictable inorganic nitrogen control.
- 2. Double protein utilization, thus enables to use cheaper feed: lower protein feed.
- 3. Lower aquaculture dependence on marine fish meal & oil.



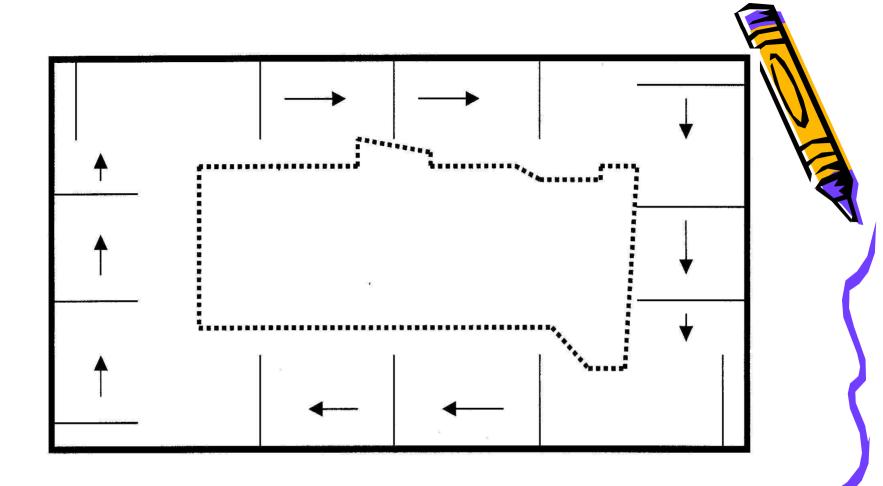


- 4. lower pollution
- 5. Slightly higher oxygen consumption.
- 6. High water turbidity, may be a problem to some species.
 - 7. Probiotic effects

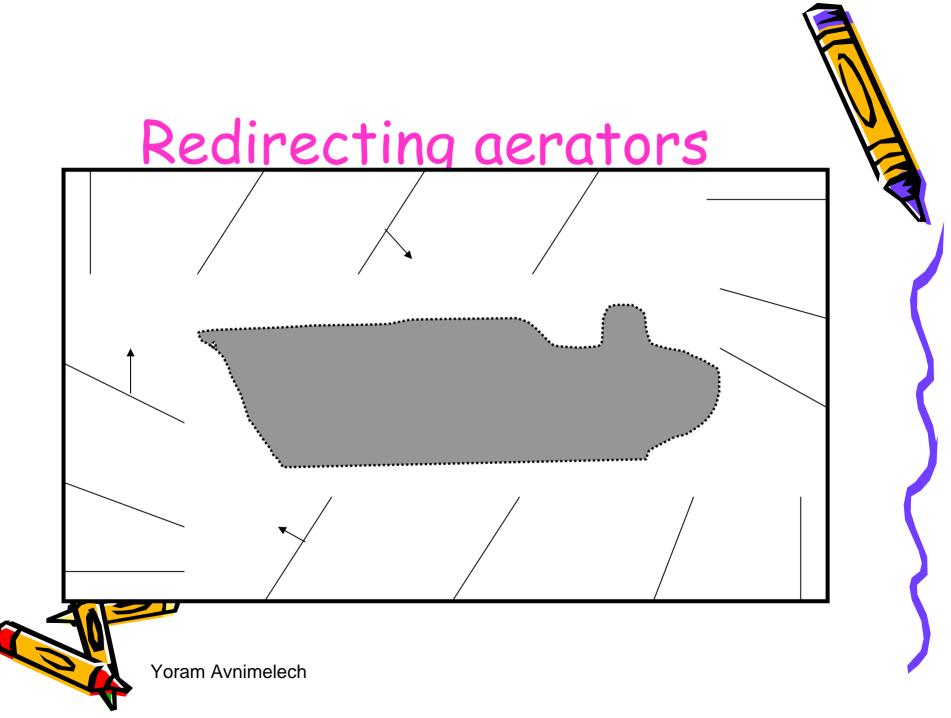
How to do it??

- 1. Enough aeration to maintain oxygen above 4-5 mg/l.
- 2. Lined pond: Plastics, concrete, soil concrete, laterite.
- 3. Placement of aerators in a way that all pond volume will be mixed.
 NO ACCUMULATION OF SLUDGE!!

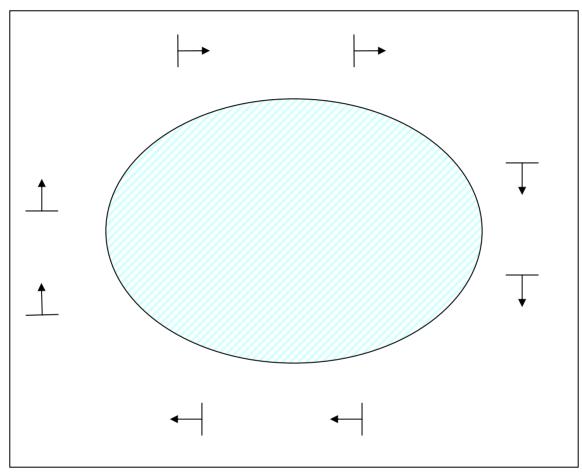




Stagnant area (Water velocity < 1 cm/sec) in 1.2 ha shrimp pond equipped with long arm paddle wheel aerators (2 hp each)

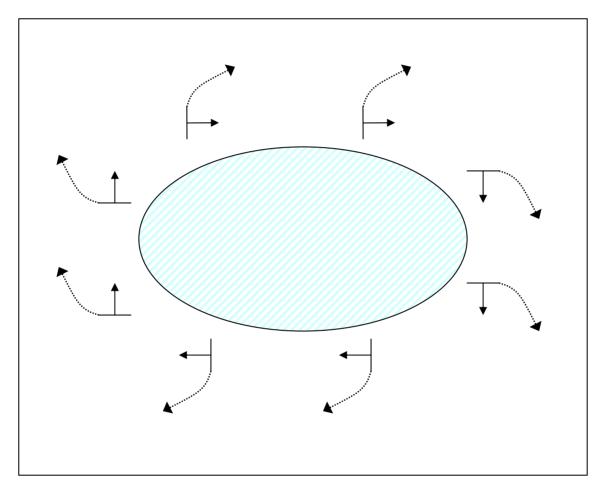


From conventional to active



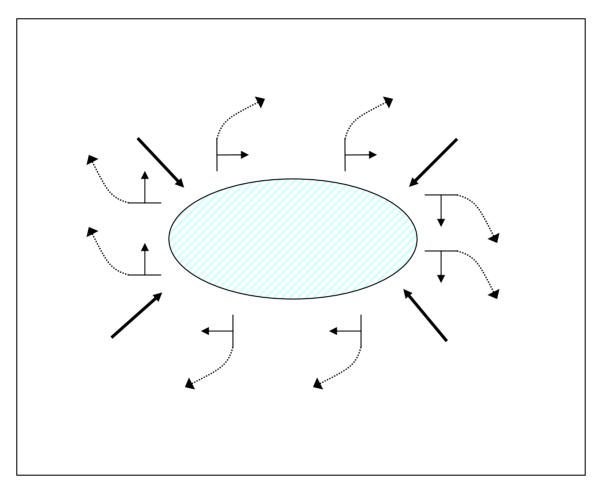


From conventional to active





From conventional to active





How to do it (2)

- 4. Feed with low protein % (20%) or add enough carbon (molasses, starch, cassawa etc.)
- 5. If inorganic nitrogen accumulates, add carbohydrates at a rate of 20 kg per kg N you want to remove.
- 6. Maintain alkalinity > 50-100 mg.



An Example: Over-wintering of tilapia

The Problem:

- One of the problems of growing tilapia in temperate regions is over-wintering. You need the whole year to complete the growing cycle of tilapia, yet, tilapia is sensitive to cold temperature and below ~ 13°C mortality starts.
- In Israel, normally it is possible to hold juveniles in ponds without catastrophes, but if cold spells occur, mortality takes place and next year there are much less than in the market.

Similar situation was report from China, where production declined by ~80% due to the cold winter 2008





Potential solution

- It is possible to keep juveniles and market size fish in green houses or similar structures during the winter, usually using solar heating, but in cases geothermal water.
- These structures are expensive. A dense biomass has to be held, in order to justify the investment. In case of dense biomass, metabolites, especially ammonium may accumulate and endanger the fish population.

Water replacement is not a reasonable option, — since you loose heat by releasing the warm water.



Materials and methods

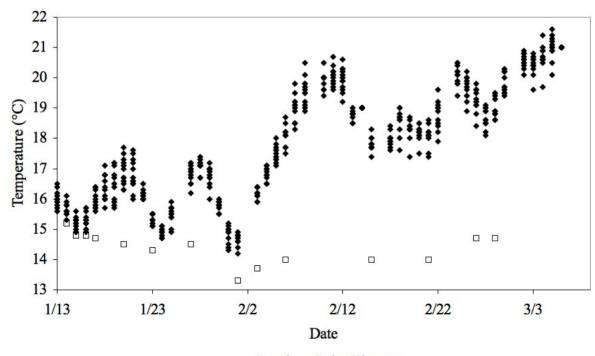
- OExperimental design
 - OWinter period 2008 (13th of January till 4th March)
 - 050 and 100 g tilapia hybrid fingerlings (*Oreochromis niloticus* x *Oreochromis aureus*)
 - 016 kg fish/m³ pond water (could be raised!)



Results

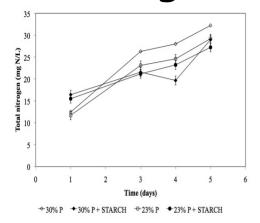
Temperature and oxygeno control

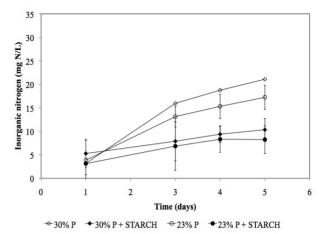
Dissolved oxygen: $9 - 10 \text{ mg } O_2/LO$ Average temperature: $18^{\circ}CO$

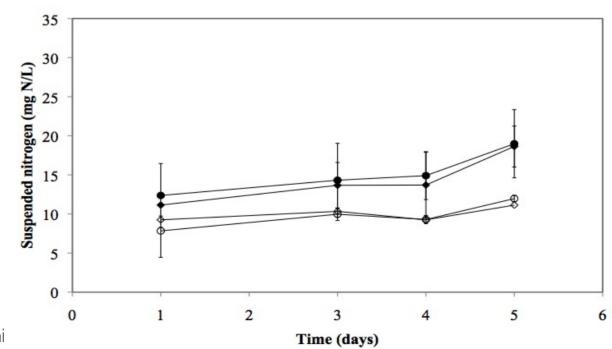




ONitrogen dynamics





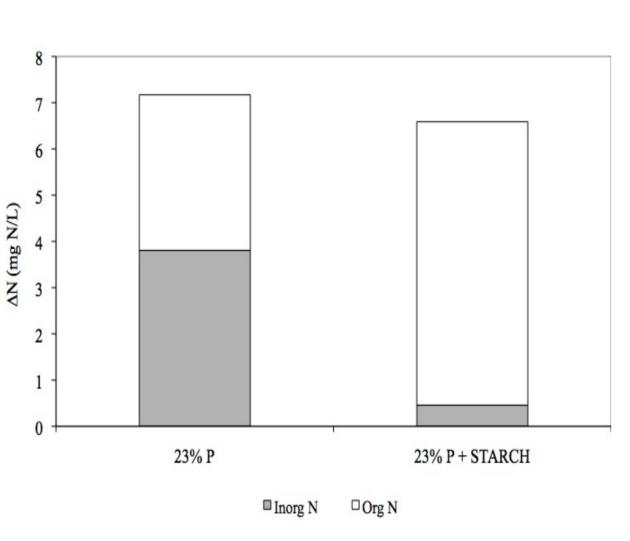


→ 30% P + STARCH

→23% P

◆ 23% P + STARCH







Fish Responses

OFish survival

○50 g fish: 80 ±4%

0100 g fish: 97 + 6%

OFish growth

 $050 g fish: 0.27 \pm 0.02$

g/fish.day

 0.29 ± 0.03

g/fish.day

OCondition factor, CF = 100.W/L

○ CF < 1.8 poor conditions

O CF > 2 good physiological state

2.17 <u>+</u> 0.06

50 g fi

2.19 + 0.07

100 g f





How to do it (3)

- 7. Minimize water exchange.
- 8. If sludge accumulates, drain sludge out or dry/clean between seasons.
- 9. It was demonstrated that the same principles can be used in stagnant ponds!!!
- Every farm is some what different.

 Learn from yours and others

 Learnience.



- Assume 1 kg shrimp/m3
- 3% feed per day = 30g
- 40% protein = 12g
- 15.5% N in protein = 1.86 g N
- 75% is released to the water ~
- ~ 1.4 g N/m3 added daily to the water (= 1.4 ppm per day)



MICROBIAL CONVERSIO

•MICROBES PRODUCE NEW CELL MATERIAL (protein) AND ENERGY:

$$\Delta C = CO_2 + \Delta C cell$$

 $\Delta C cell/\Delta C = \varepsilon$

- = Microbial conversion efficiency
- =normally, 0.4-0.6 for aerobic microbial processes. Lower for anaerobic.
- Bacteria are rich in respect to N (C:N ~ 4)

P Nitrogen is taken up for $4 \Delta C$