

**Natural enrichment of ^{15}N as a
tool to quantitatively evaluate
protein uptake
in biofloc systems.**

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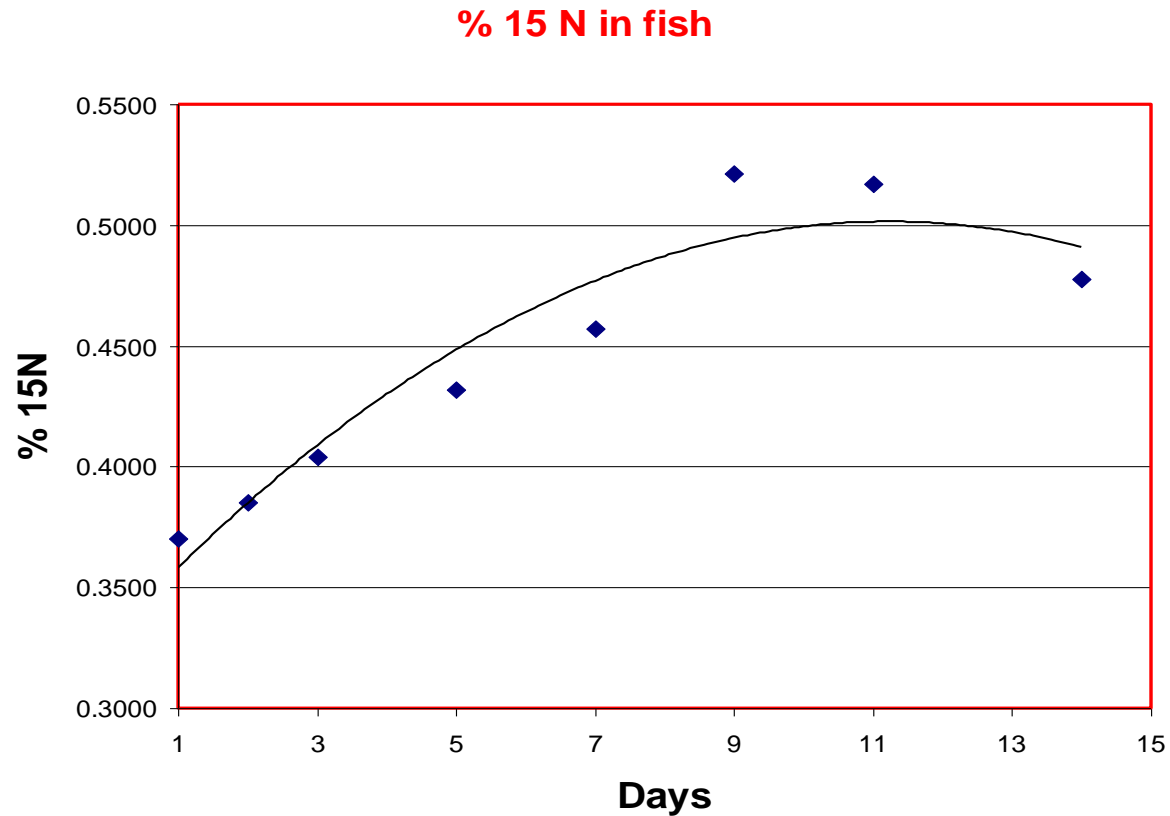
What do our fish (shrimp) really eat?

It is hard to know
They are under the water

It is possible to use enrichment of either the feed pellets or the bio-sources with ^{15}N . By adding $^{15}\text{NH}_4\text{Cl}$ (or other enriched ammonium salt) to a biofloc suspension, the bioflocs are tagged and their uptake by the target animal can be determined.

Michele Burton and coworkers (e.g. *Aquaculture* 232:525-537, 2004) used this method to follow protein uptake by shrimp and Avnimelech (e.g. *Aquaculture* 287:163-168, 2008)

Results 1: % ^{15}N in fish



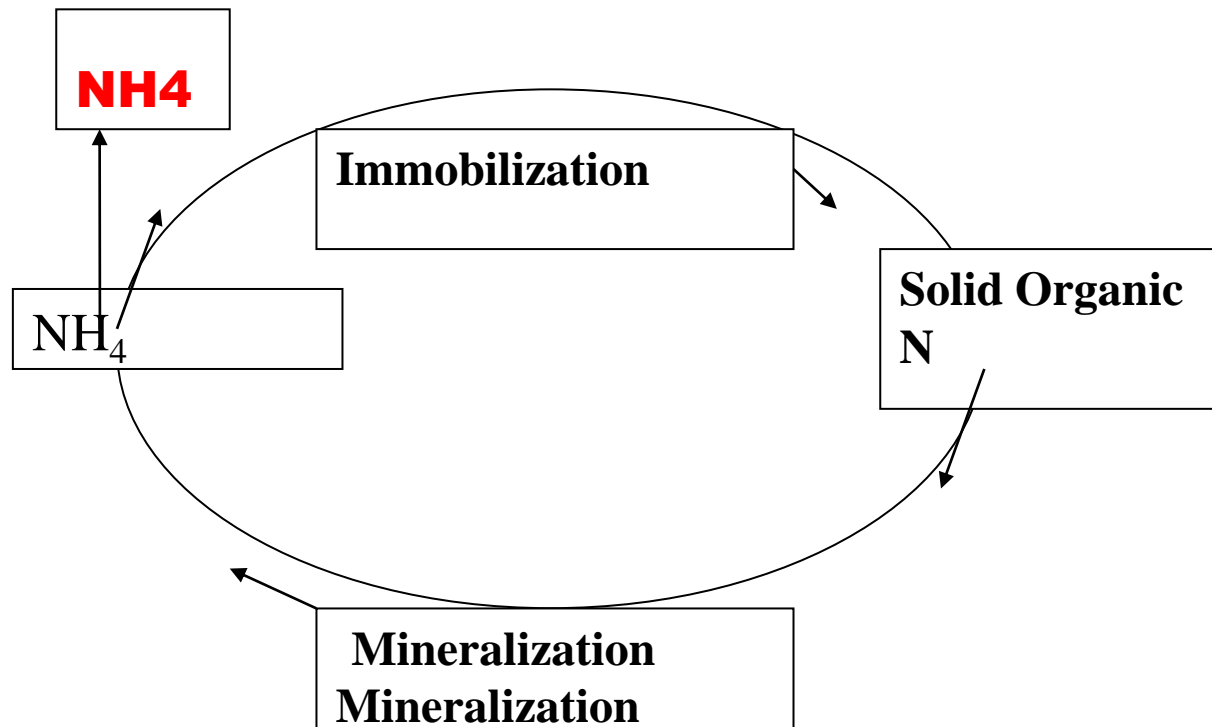
It is not too difficult to do

- The determination of ^{15}N enrichment is rather fast and not exceedingly expensive, using automated IRMS (Isotope Ratio Mass Spectrometers) available in quite a few centers.
- **BUT:**
- The enriched ^{15}N is expensive ~ \$100/g
- Thus, we can use it only in small systems, not in real ponds.

Nitrogen is naturally made of 2 stable (non radioactive) isotopes. The common (99.64%), light isotope is ^{14}N , while 0.36% of nitrogen is ^{15}N .

The two isotopes are chemically identical, yet, when nitrogen undergoes a set of reactions, the products of the reaction may have different isotopic ratios as compared to the source materials. (This differentiation may be due to somewhat stronger attraction of the heavier isotope or to faster diffusion of the lighter isotope).

Natural enrichment



Results, Tilapia

Table 1. Preliminary test of ^{15}N & ^{13}C in pond components

Tilapia BFT pond, Jordan Valley, Israel

Sample	$\delta^{15}\text{N}$ % (*)	^{13}C % (*)
Feed pellets	0.27	-18.4
Bioflocs	1.02	- 20.9
Fish tissue	0.63	-21.7

(*) % deviation from global isotope ratio standards



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Conclusions

- **^{15}N in the biofloc was way higher than in the feed (the only source of nitrogen in the pond). The ^{15}N enrichment ratio in the fish, was intermediate between its enrichment in the feed and in the bioflocs. With the values obtained we can assume that **48% of the nitrogen in the fish came from the bioflocs.****

Vertical Substrates, Hitide Sea Farms, India, Sept. 2011



Results in semi-intensive shrimp pond in India

- Table 3 ^{15}N enrichment in a biofloc shrimp pond in India

• Sample	Feed pellets	Biofloc	Periphyton	Shrimp
□ $\delta^{15}\text{N}$	0.59	1.3	1.58	7.3

Conclusions

- Our preliminary results show that we can use natural enrichment to tag bioflocs and periphyton.
- This is a powerful and affordable method to determine uptake of natural feed sources by fish and shrimp.
- This technology should be further tested and developed.