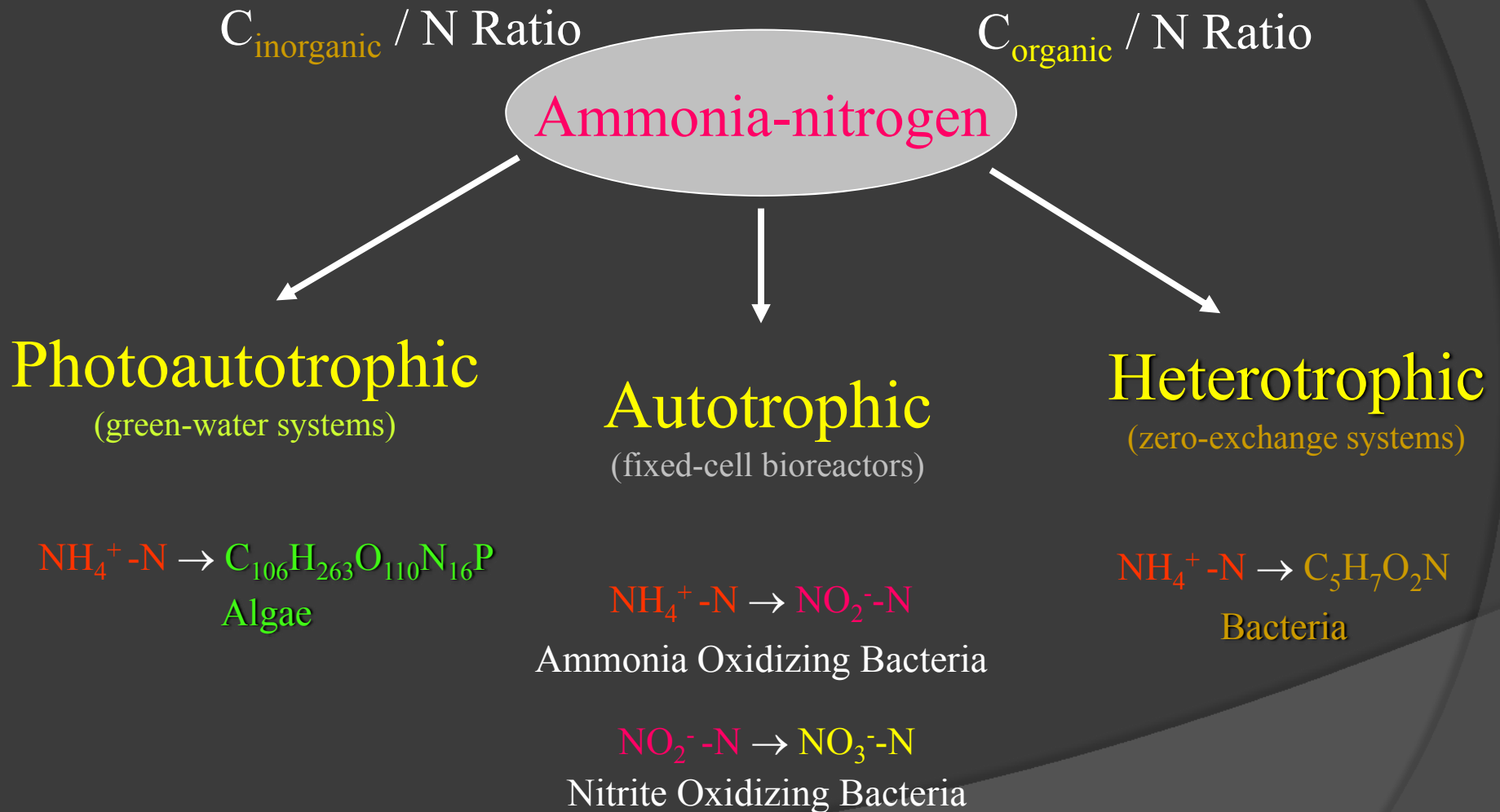


# THE THREE PATHWAYS FOR THE REMOVAL OF AMMONIA-NITROGEN IN AQUACULTURE SYSTEMS

James M. Ebeling, Ph.D.  
Research Engineer  
Aquaculture Systems Technologies, LLC

Michael B. Timmons, Ph.D.  
Professor  
Dept. of Bio. & Environ. Eng.  
Cornell University

# Introduction



# “New Paradigm” → ????

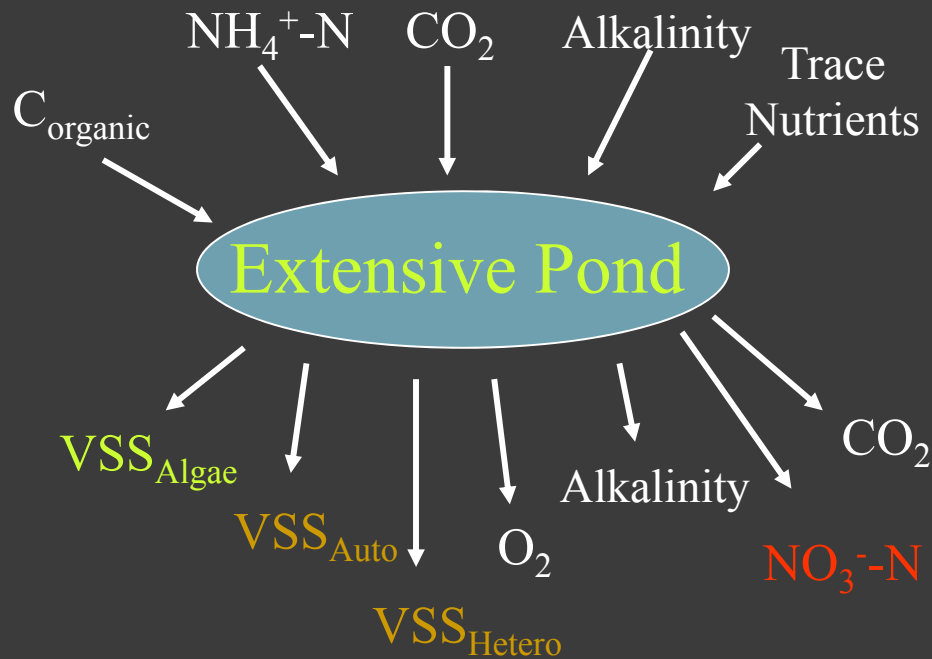
## ??? Understanding of the ‘Removal System’

- Photoautotrophic
- Autotrophic
- Heterotrophic
- **Some Combination!**

**Impact on Water Quality!!!!**

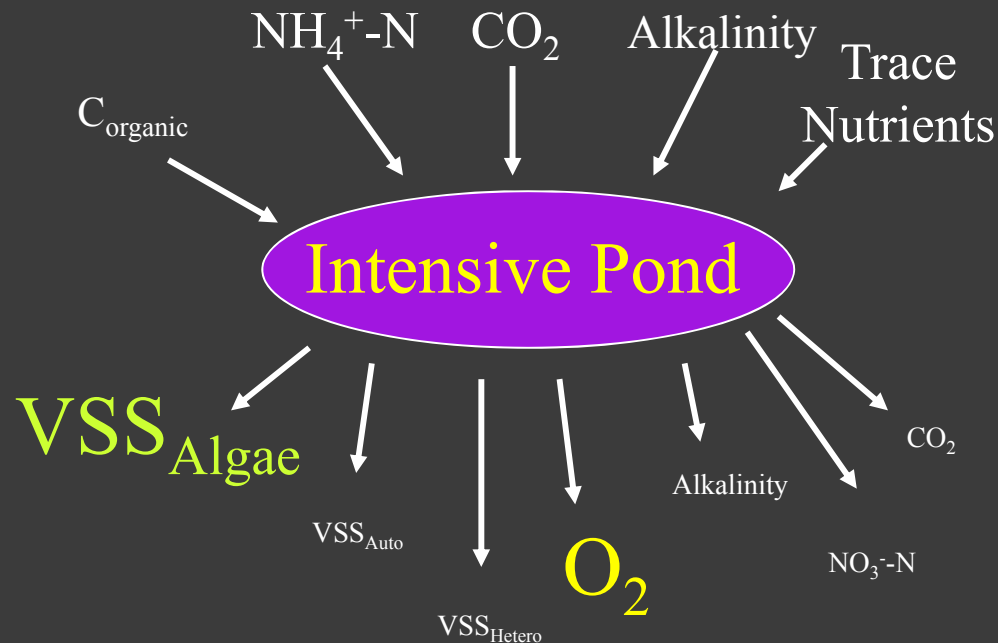
**Management Strategies!**

# Nitrogen Removal Pathways



- Photoautotrophic
- Autotrophic
- Heterotrophic
- *Other Mysterious Ways*

# Nitrogen Removal Pathways

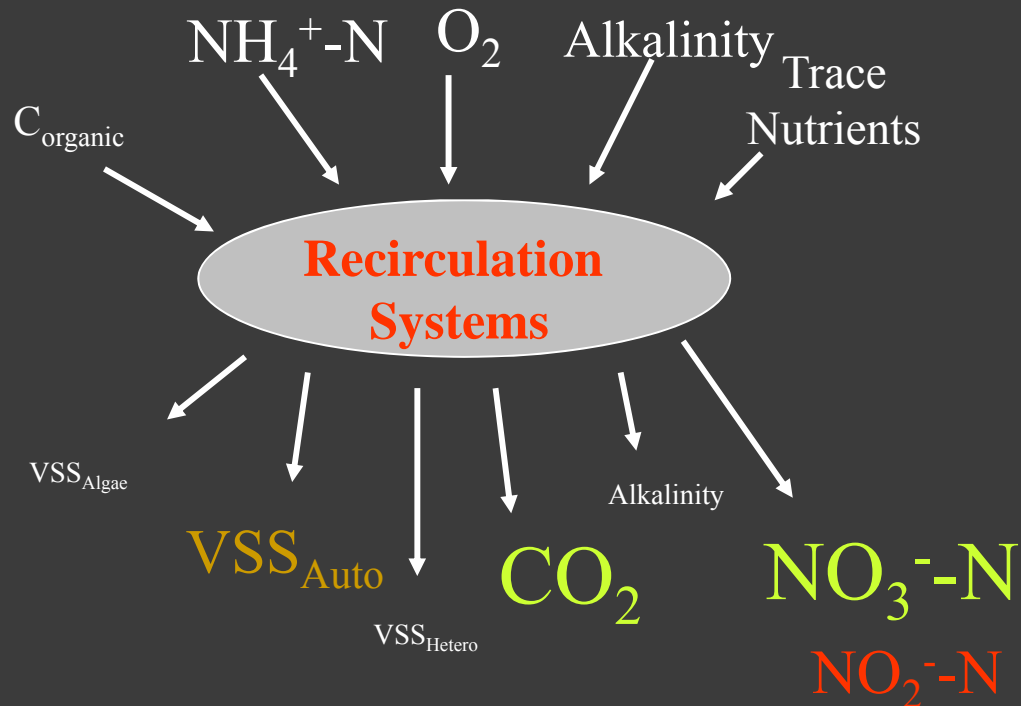


## • Photoautotrophic

- Autotrophic
- Heterotrophic
- Other Mysterious Ways

Algae Based Systems

# Nitrogen Removal Pathways



- Photoautotrophic

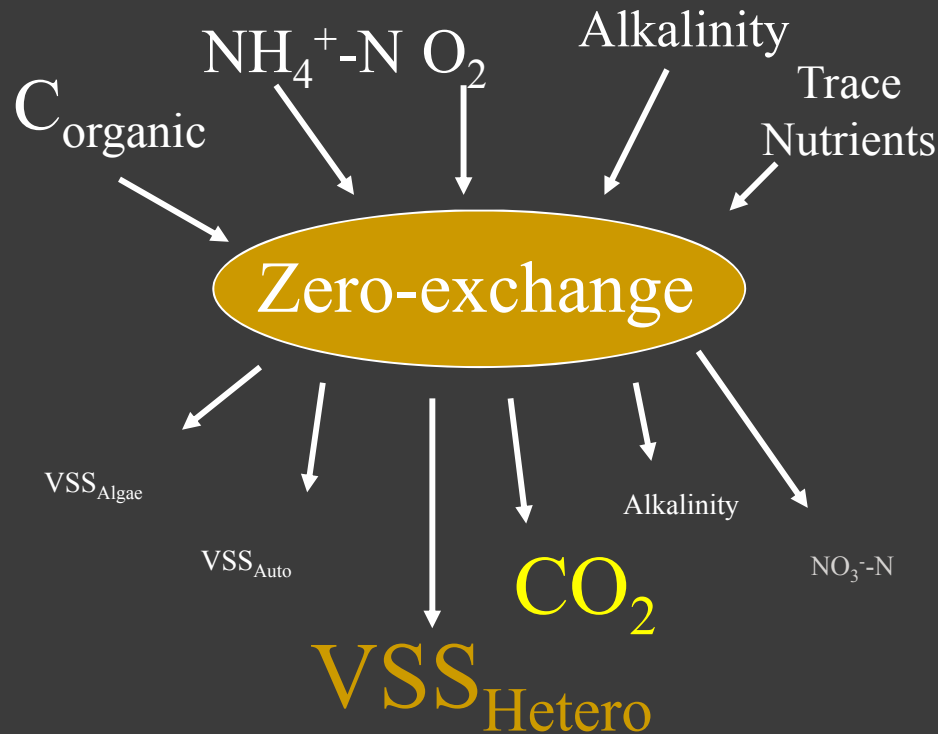
- **Autotrophic**

- Heterotrophic

- Denitrification

Fixed-film Bioreactors

# Nitrogen Removal Pathways



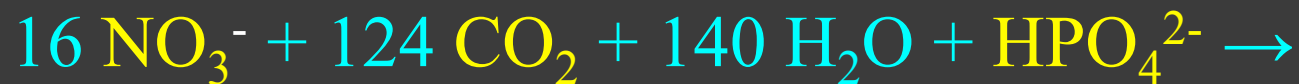
- Photoautotrophic
- Autotrophic
- **Heterotrophic**
- Denitrification

Suspended Growth Systems

# Photoautotrophic (algal based systems)

## Biosynthesis of saltwater algae:

Nitrate as nitrogen source



Ammonia as nitrogen source





# Photoautotrophic (algal based systems)

		Consumes	C <sub>organic</sub>	C <sub>inorganic</sub>	N
Consumables	Stoichiometry	(g)	(g)	(g)	(g)
NH <sub>4</sub> <sup>+</sup> -N		1.0	-----	-----	1.0
Carbon Dioxide	18.07 g CO <sub>2</sub> / g N	18.07	-----	4.93	-----
Alkalinity	3.13 g Alk/ g N	3.13	-----	0.75	-----
		Yields	C <sub>organic</sub>	C <sub>inorganic</sub>	N
Products	Stoichiometry	(g)	(g)	(g)	(g)
VSS <sub>Algae</sub>	15.85 g VSS <sub>A</sub> / g N	15.85	5.67	-----	1.0
Oxygen	15.14 g O <sub>2</sub> / g N	15.14	-----	-----	-----

# Autotrophic - Nitrification

## Biosynthesis of Autotrophic bacteria:



The major factors affecting the rate of nitrification include:

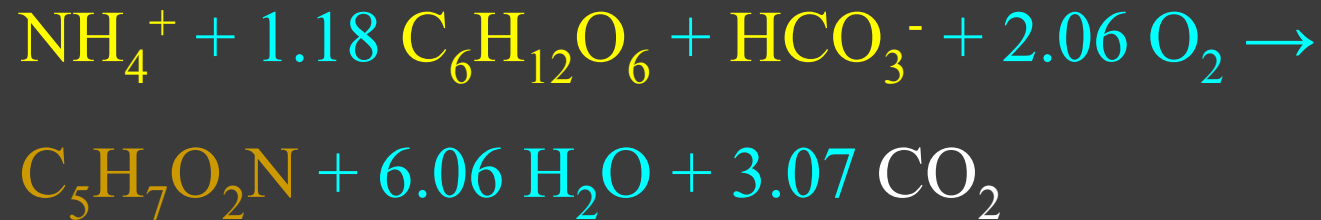
- ammonia-nitrogen and nitrite-nitrogen concentration
- carbon/nitrogen ratio
- dissolved oxygen
- pH
- temperature
- alkalinity
- salinity

# Autotrophic - Nitrification

		Consumes	C <sub>organic</sub>	C <sub>inorganic</sub>	N
Consumables	Stoichiometry	(g)	(g)	(g)	(g)
NH <sub>4</sub> <sup>+</sup> -N		1.0	-----	-----	1.0
Alkalinity	7.05 g Alk/ g N	7.05	-----	1.69	-----
Oxygen	4.18 g O <sub>2</sub> / g N	4.18	-----	-----	-----
		Yields	C <sub>organic</sub>	C <sub>inorganic</sub>	N
Products	Stoichiometry	(g)	(g)	(g)	(g)
VSS <sub>A</sub>	0.20 g VSS <sub>A</sub> / g N	0.20	0.11	-----	0.024
NO <sub>3</sub> <sup>-</sup> -N	0.976 g NO <sub>3</sub> <sup>-</sup> -N /g N	0.976	-----	-----	0.976
CO <sub>2</sub>	5.85 g CO <sub>2</sub> / g N	5.85	-----	1.58	-----

# Heterotrophic Bacteria

## Biosynthesis of Heterotrophic bacteria:



The major factors affecting the rate of nitrification include:

- ammonia-nitrogen
- carbon/nitrogen ratio
- dissolved oxygen
- pH
- temperature
- alkalinity
- salinity

# Heterotrophic Bacteria

		Consumes	C <sub>organic</sub>	C <sub>inorganic</sub>	N
Consumables	Stoichiometry	(g)	(g)	(g)	(g)
NH <sub>4</sub> <sup>+</sup> -N		1.0	-----	-----	1.0
C <sub>6</sub> H <sub>12</sub> O <sub>6</sub>	15.17 g Carbs/ g N	15.17	6.07	-----	-----
Alkalinity	3.57 g Alk/ g N	3.57	-----	0.86	-----
Oxygen	4.71 g O <sub>2</sub> / g N	4.71	-----	-----	-----
		Yield	C <sub>organic</sub>	C <sub>inorganic</sub>	N
Products	Stoichiometry	(g)	(g)	(g)	(g)
VSS <sub>H</sub>	8.07 g VSS <sub>H</sub> / g N	8.07	4.29	-----	1.0
CO <sub>2</sub>	9.65 g CO <sub>2</sub> / g N	9.65	-----	2.63	-----

# Impact of C/N Ratio

↓ **C/N Ratio** ↑

$C_{\text{labile}}/N \sim 0$   
 $C_{\text{organic}}/N \sim \text{small}$

$C_{\text{labile}}/N \sim 2.2$   
 $C/N \sim 8-10$

$C_{\text{labile}}/N \sim 6.2$   
 $C/N \sim 12-16$

Autotrophic ←  Heterotrophic

inorganic carbon  
as alkalinity

organic carbon  
from the feed

organic carbon from the feed  
plus supplemental carbohydrates

100 % Autotrophic

(109 g  $C_{\text{organic}}$  /kg feed)  
@ 35% protein

100% Heterotrophic

(Recirculation System with  
excellent solids removal)

56 % Heterotrophic  
44 % Autotrophic

# Autotrophic (Intensive Recirculation System)

Autotrophic System

Conversion of 1 kg of feed @ 35% protein

$$32.2 \text{ g N} * 0.20 \text{ g VSS/ g N}$$

$$= 6.44 \text{ g VSS}_{\text{autotrophic}}$$

$$0.124 \text{ gN/gVSS}_A$$

$$0.531 \text{ gC/gVSS}_A$$

$$0.80 \text{ g N}_{\text{VSS}}$$

$$3.42 \text{ g C}_{\text{VSS}}$$

$$+ 31.4 \text{ g NO}_3\text{-N}$$

$$+ 51.3 \text{ g CO}_2$$

# Autotrophic (Intensive Recirculation System)

		Consumes	C <sub>organic</sub>	C <sub>inorganic</sub>	N
Consumables	Stoichiometry	(g)	(g)	(g)	(g)
NH <sub>4</sub> <sup>+</sup> -N		32.2	-----	-----	32.2
Alkalinity	7.05 g Alk/ g N	227	-----	54.7	-----
Oxygen	4.18 g O <sub>2</sub> / g N	135	-----	-----	-----
		Yields	C <sub>organic</sub>	C <sub>inorganic</sub>	N
Products	Stoichiometry	(g)	(g)	(g)	(g)
VSS <sub>A</sub>	0.20 g VSS <sub>A</sub> / g N	6.44	3.4	-----	0.8
NO <sub>3</sub> <sup>-</sup> -N	0.976 g NO <sub>3</sub> <sup>-</sup> -N / g N	31.4	-----	-----	31.4
CO <sub>2</sub>	5.85 g CO <sub>2</sub> / g N	188	-----	51.3	-----

54.7g C<sub>Alk</sub> / 32.2 g N → C/N ratio of 1.6 and TOC is very, very small



# Biofloc System (?) (no Carbon Supplementation)

## Heterotrophic and Autotrophic Components

Heterotrophic Component: Organic Carbon from Feed

$$1 \text{ kg}_{\text{feed}} * 0.36 \text{ kg BOD/kg feed} * 0.40 \text{ kg VSS/ kg BOD} =$$

$$= 144 \text{ g VSS}_{\text{heterotrophic}}$$

$$0.124 \text{ gN/gVSS}_{\text{H}}$$

$$0.531 \text{ gC/gVSS}_{\text{H}}$$

$$17.9 \text{ g N}_{\text{VSS}}$$

$$76.5 \text{ g C}_{\text{VSS}}$$

$$+ 47.1 \text{ g C}_{\text{CO}_2} = 123.6 \text{ g}$$

C

$$109 \text{ g C}_{\text{feed}}$$

$$15.4 \text{ g C}_{\text{Alkalinity}}$$

Assume that the heterotrophic bacteria out compete the autotrophic bacteria.

# Biofloc System (?) (no Carbon Supplementation)

## Heterotrophic and Autotrophic Components

### Autotrophic Component: Inorganic Carbon Alkalinity

Excess Ammonia-nitrogen:

$$32.2 \text{ g NH}_3\text{-N} - 17.9 \text{ g N}_{\text{VSS}} = 14.3 \text{ g N}_A$$

$$14.3 \text{ g N} * 0.20 \text{ g VSS/ g N}$$

$$= 2.86 \text{ g VSS}_{\text{autotrophic}}$$

$$0.124 \text{ gN/gVSS}_A$$

$$0.531 \text{ gC/gVSS}_A$$

$$0.35 \text{ g N}_{\text{VSS}}$$

$$1.52 \text{ g C}_{\text{VSS}}$$

$$+ 14 \text{ g NO}_3\text{-N}$$

$$+ 24.3 \text{ g C}_{\text{Alk}}$$

# Biofloc System (?) (no Carbon Supplementation)

## Heterotrophic and Autotrophic Components

### *Heterotrophic Bacteria*

	Stoichiometry	Consumes (g)	C <sub>organic</sub> (g)	C <sub>inorganic</sub> (g)	N (g)
NH <sub>4</sub> <sup>+</sup> -N	0.56 * N <sub>T</sub>	17.9	-----	-----	17.9
C <sub>6</sub> H <sub>12</sub> O <sub>6</sub> feed	15.17 g Carbs/ g N	272	109	-----	-----
Alkalinity	3.57 g Alk/ g N	63.9	-----	15.4	-----

### *Autotrophic Bacteria*

	Stoichiometry	Consumes (g)	C <sub>organic</sub> (g)	C <sub>inorganic</sub> (g)	N (g)
NH <sub>4</sub> <sup>+</sup> -N	0.44 * N <sub>T</sub>	14.3	-----	-----	14.3
Alkalinity	7.05 g Alk/ g N	101	-----	24.3	-----

Conversion of 1 kg of feed @ 35% protein

# Biofloc System (?) (no Carbon Supplementation)

## Heterotrophic and Autotrophic Components

### *Heterotrophic Bacteria*

		Yields	C <sub>organic</sub>	C <sub>inorganic</sub>	N
	Stoichiometry	(g)	(g)	(g)	(g)
VSS <sub>H</sub>	8.07 g VSS <sub>H</sub> / g N	144	76.5	-----	17.9
CO <sub>2</sub>	9.65 g CO <sub>2</sub> / g N	173	-----	47.1	-----

### *Autotrophic Bacteria*

		Yields	C <sub>organic</sub>	C <sub>inorganic</sub>	N
	Stoichiometry	(g)	(g)	(g)	(g)
VSS <sub>A</sub>	0.20 g VSS <sub>A</sub> / g N	2.86	3.45	-----	0.35
NO <sub>3</sub> <sup>-</sup> -N	0.976 g NO <sub>3</sub> <sup>-</sup> -N/g N	31.7	-----	-----	13.9
CO <sub>2</sub>	5.85 g CO <sub>2</sub> / g N	83.4	-----	22.8	-----

Conversion of 1 kg of feed @ 35% protein

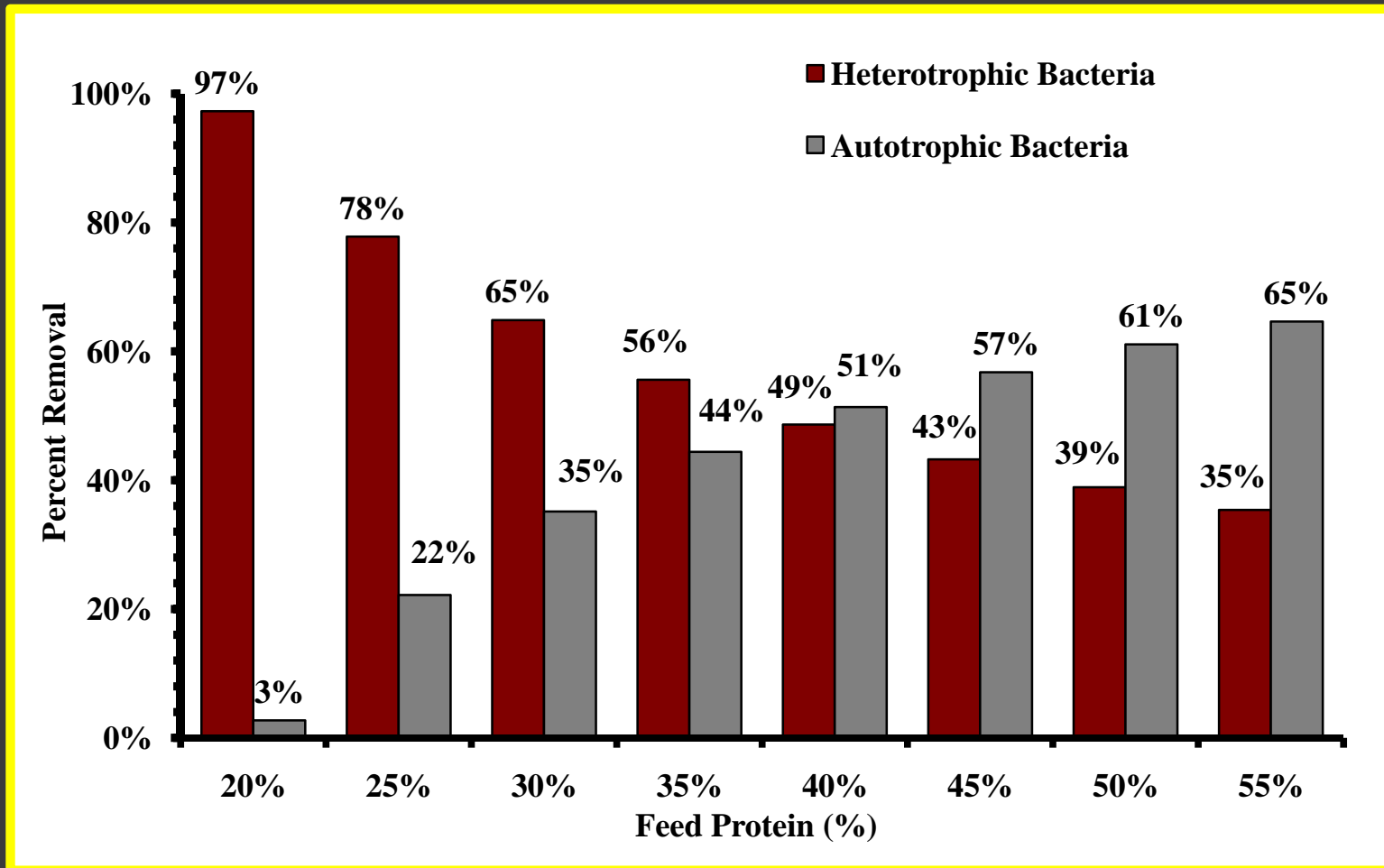
# Biofloc System (?) (no Carbon Supplementation)

## Heterotrophic and Autotrophic Components

Consumes		C <sub>organic</sub> (g)	C <sub>inorganic</sub> (g)	N (g)
NH <sub>4</sub> <sup>+</sup> -N	32.2 g N	-----	-----	32.2
C <sub>6</sub> H <sub>12</sub> O <sub>6</sub>	272 g Carbs (feed)	109	-----	-----
Alkalinity	165 g Alk	-----	40	-----
Yields		C <sub>organic</sub> (g)	C <sub>inorganic</sub> (g)	N (g)
VSS	147 g VSS	80	-----	18.2
NO <sub>3</sub> <sup>-</sup> -N	31.7 g NO <sub>3</sub> <sup>-</sup> -N	-----	-----	13.9
CO <sub>2</sub>	256 g CO <sub>2</sub>	-----	70	-----

Conversion of 1 kg of feed @ 35% protein

# Biofloc System (?) (no Carbon Supplementation) Heterotrophic and Autotrophic Components



Percent removal of ammonia-nitrogen by Heterotrophic or Autotrophic Process  
as a function of % Protein

# Biofloc System

## Carbon Supplement

Excess Ammonia-nitrogen:

$$32.2 \text{ g NH}_3\text{-N} - 17.9 \text{ g N}_{\text{VSS}} = 14.3 \text{ g N}_A$$

$$14.3 \text{ g N} * 8.07 \text{ g VSS} / \text{g N}$$

$$= 115 \text{ g VSS}_{\text{heterotrophic}}$$

$$0.124 \text{ gN/gVSS}_H$$

$$0.531 \text{ gC/gVSS}_H$$

$$14.3 \text{ g N}_{\text{VSS}}$$

$$61.1 \text{ g C}_{\text{VSS}}$$

$$+ 37.6 \text{ g C}_{\text{CO}_2} = 98.7 \text{ g C}$$

$$86.4 \text{ g C}_s$$

$$12.3 \text{ g C}_{\text{Alkalinity}}$$

Carbohydrate is 40% Carbon  $\Rightarrow$  216 g carbs

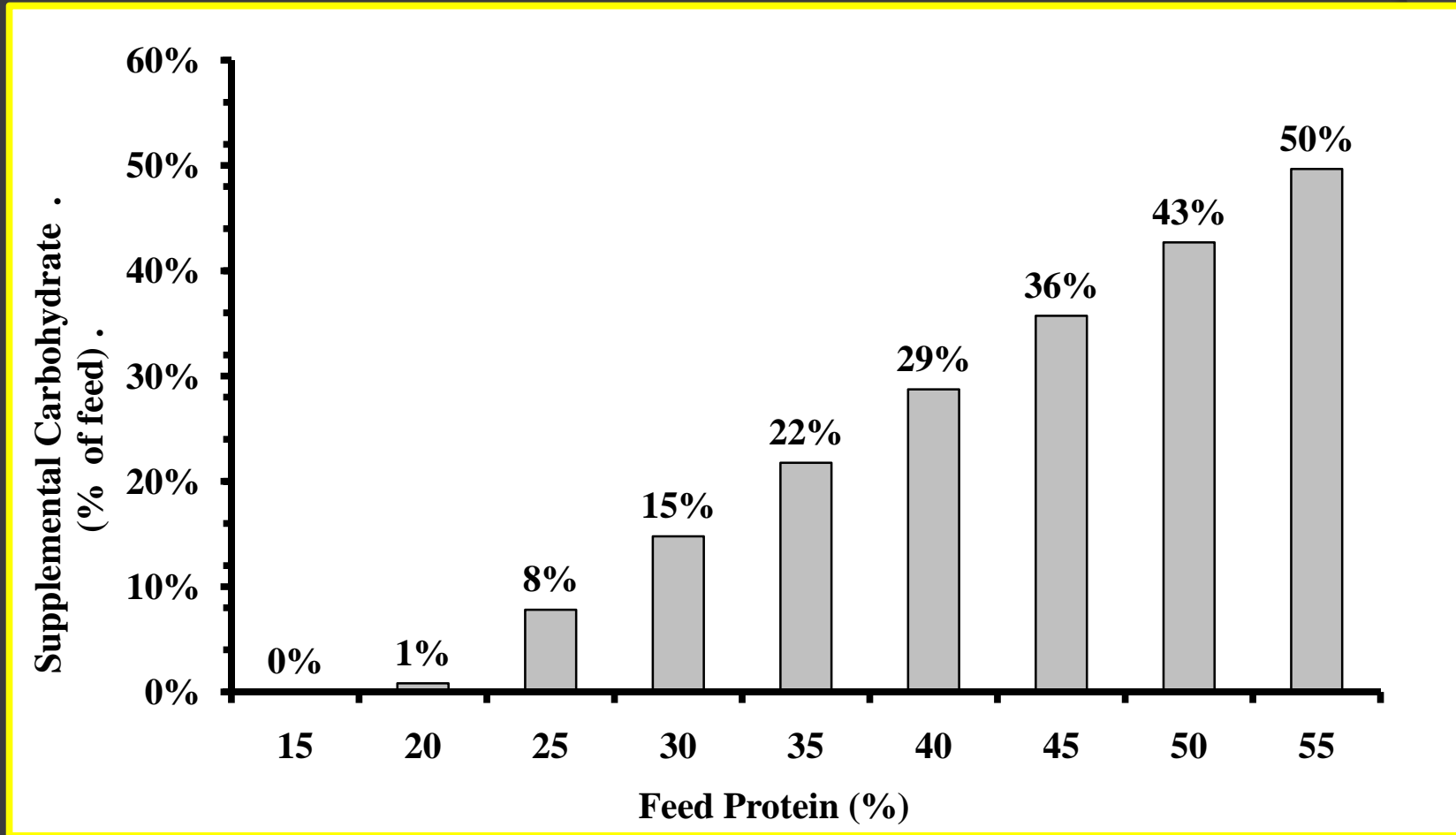
# Biofloc System (Carbon Supplementation)

		Consumes	C <sub>organic</sub>	C <sub>inorganic</sub>	N
Consumables	Stoichiometry	(g)	(g)	(g)	(g)
NH <sub>4</sub> <sup>+</sup> -N		32.2	-----	-----	32.2
C <sub>6</sub> H <sub>12</sub> O <sub>6</sub>	15.17 g Carbs/ g N	488	196	-----	-----
Alkalinity	3.57 g Alk/ g N	115	-----	27.7	-----
Oxygen	4.71 g O <sub>2</sub> / g N	152	-----	-----	-----
		Yield	C <sub>organic</sub>	C <sub>inorganic</sub>	N
Products	Stoichiometry	(g)	(g)	(g)	(g)
VSS <sub>H</sub>	8.07 g VSS <sub>H</sub> / g N	260	138	-----	32.2
CO <sub>2</sub>	9.65 g CO <sub>2</sub> / g N	311	-----	85	-----

Conversion of 1 kg of feed @ 35% protein

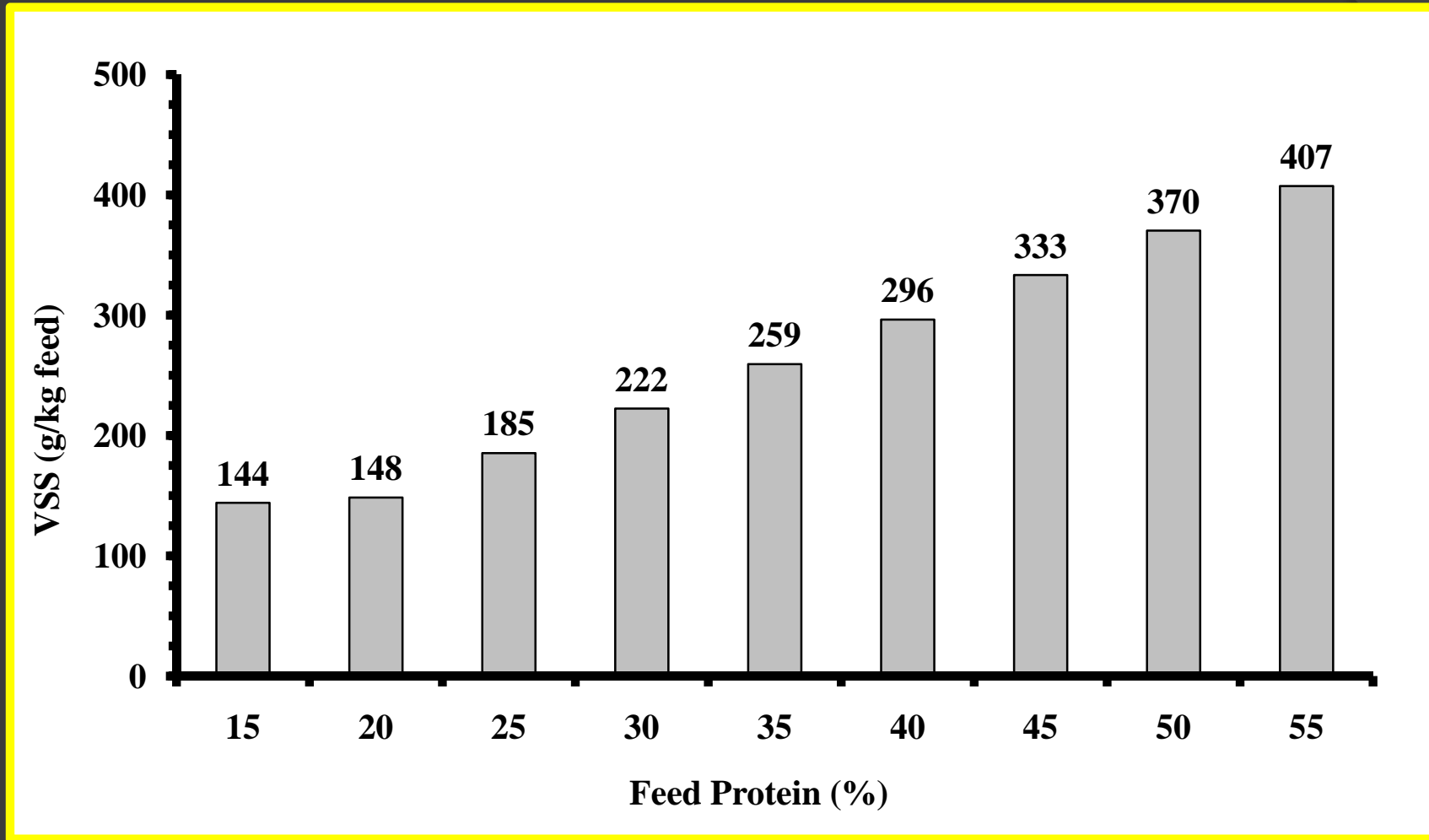


# Biofloc System (Carbon Supplementation)



Supplemental Carbohydrate as percentage of feed rate  
for heterotrophic metabolism of ammonia-nitrogen to microbial biomass

# Biofloc System (Carbon Supplementation)



VSS production as percentage of feed rate  
for heterotrophic metabolism of ammonia-nitrogen to microbial biomass

# Conclusions

“Engineering Sustainability”

Organic Carbon + Nitrogen → Bacterial Biomass



# Questions?

