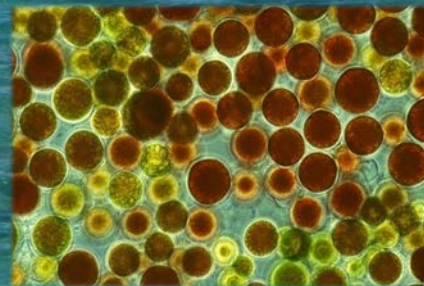


Evaluation of Biofuel Co-products as Ingredients for Aquafeeds

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Biofuel Co-products



Oil Seeds and Nuts
Algae, Coconut, Jatropha, Rapeseed, Camelina



Processing
Clean Crack Extrude Press



Oil

Biodiesel



By-products

Feeds

Outline

- **Criteria and methodology for evaluation of an ingredient**
- **Substitution of fishmeal protein by biofuel co-products in aquafeeds (opportunity and concerns)**
- **Discussion on future researches**

Essential Information for Evaluation of an Ingredient

- **Nutrient requirement of a target species**
- **Sustainability for production of the ingredient**
- **Nutritional evaluation of the ingredient**

Selection Criteria for an Ingredient

---Nutritional evaluation

- **Chemical composition**
- **Pellet physical quality**
- **Effects on attractiveness and palatability**
- **Digestibility and utilization of nutrients**
- **Effect on product quality**

Evaluation of an Ingredient

Chemical Composition

- **Nutrient levels**
- **Nutrient balance (amino acid; fatty acid)**
- **Presence of anti-nutrients or contaminants**

Proximate Composition of Different Ingredients (%)

Analysis	Fishmeal	Soybean meal	DDGS	Camelina	Algae
DM	93	92	90	94	95
Protein	68	46	28	29	32
Lipid	6.5	1.9	5.7	33.5	0.9
Ash	18.5	5.1	5.2	3.5	17.9
Crude fiber		6.6	7.0		39.5

Amino acid profiles of different ingredients (% of total amino acids)

Amino acids	Pollock meal	Menhaden	SBM	DDGS	Camelina	Algae meal
Ala	6.2	7.8	5.1	7.5	5.3	10.6
Asp+ASN	8.8	9.6	8.9	6.6	7.8	6.6
Cys	0.8	0.6	1.0	2.2	1.4	0.8
Glu+Gln	12.3	11.0	15.7	16.3	13.2	12.1
Gly	8.4	8.1	4.9	4.3	5.0	9.9
Pro	6.2	4.4	5.1	8.8	5.2	8.0
Ser	4.7	3.6	4.9	4.7	4.2	5.4
Tyr	3.8	3.5	3.5	5.6	3.7	4.0
Taurine	2.0	1.0			0.2	0.1
Arg	7.6	9.2	8.9	4.7	9.0	6.2
His	2.2	2.9	3.6	3.4	3.4	2.4
Ile	5.6	4.7	5.3	4.1	5.4	3.6
Leu	6.7	7.5	7.1	10.9	9.4	9.4
Lys	8.5	9.6	6.8	3.3	6.8	4.9
Met	2.7	2.6	1.5	2.2	2.4	1.2
Phe	4.3	4.9	7.7	5.5	7.0	6.1
Thr	4.8	4.7	4.6	3.7	4.5	6.2
Val	5.5	5.8	5.5	5.3	6.2	4.3

Compositions of Traditional Ingredients & Microalgae

Ingredients	DM	Ash	CP	EE	Fe
			%		mg/kg
Menhaden meal	91.9	20.7	62.8	9.3	739
Soybean meal	92.5	6.8	47.8	1.9	53
Chlorella A	95.9	19.4	38.2	4.0	5882
Chlorella B	97.3	22.4	31.5	3.6	1592
Haematococcus pluvialis	91.6	10.8	40.3	5.0	75

Evaluation of an Ingredient

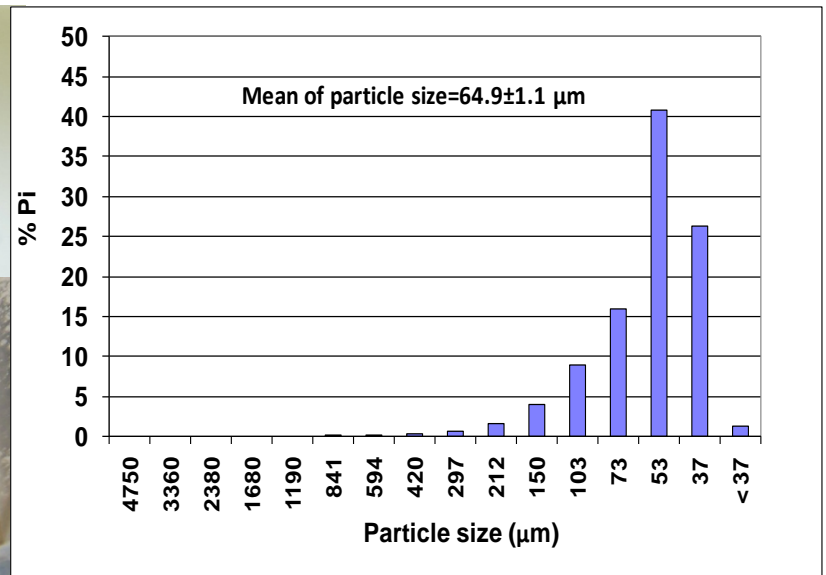
Pellet Physical Quality

- Ingredient particle sizes
- Pellet durability
- Expansion of pellets after extrusion
- Oil absorption capacity of pellets
- Pellet water stability



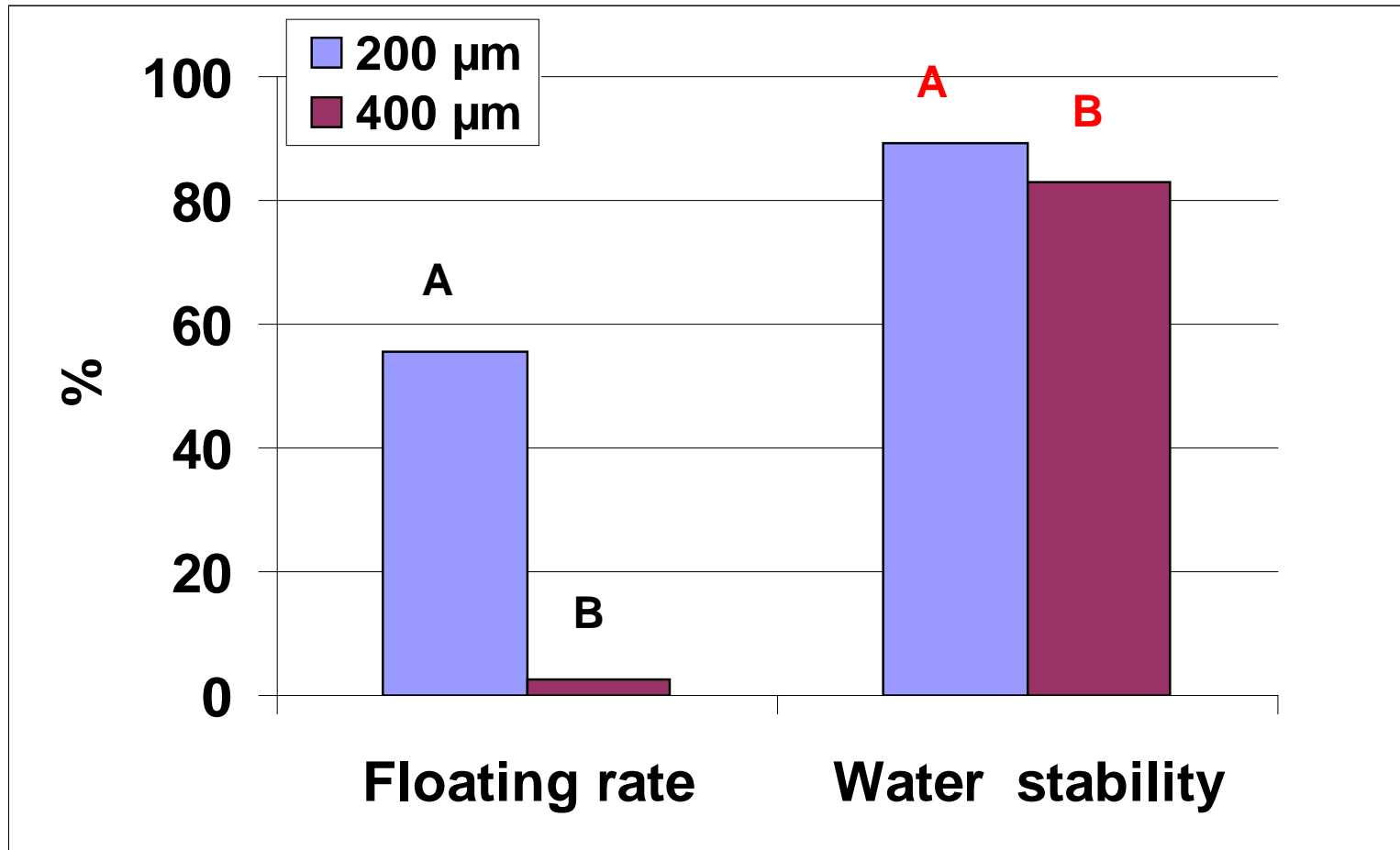
Particle Size of Ingredients

- **Physical parameters of pellets**
homogeneity, density and water stability
- **Nutrient utilization**



Particle size < 200 μm

Effect of Different Particle Size on Floating and Water Stability of Pellets



Pellet Durability



Tumble pellets & 5-½” hex nuts @10 minutes at 50 rpm



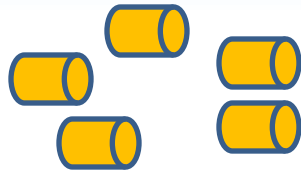
Sieve and remove the fine particles



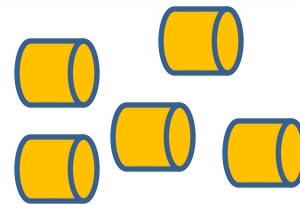
PD (%) = 100 * pellet after tumbling (g) / pellet before tumbling (g)

Pellet Expansion Measurement

Base diet



70% base diet + 30% test ingredient



Expansion: % change in the diameter of the test pellet

$$= ((\text{pellet width} - \text{die diameter}) \times \text{die diameter}^{-1}) \times 100$$

Pellet Characteristic & Density

Pellet characteristic	Sea water @ 20°C	Fresh water @ 20°C
	Density (g/L)	
Fast sinking	>640	>600
Slow sinking	580-600	540-560
Neutral	520-540	480-520
Floating	<480	<440

Oil Absorption Capacity

Weigh pellet sample (W1)



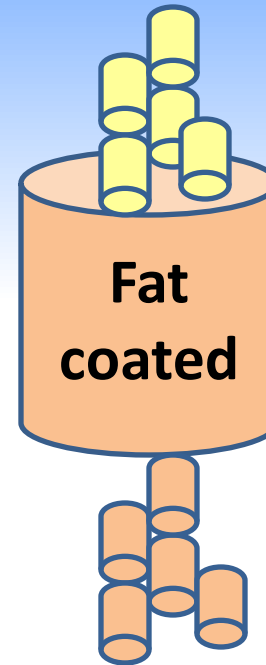
Fat coated pellets



Drain pellets for 10 mins



Weigh pellet sample (W2)



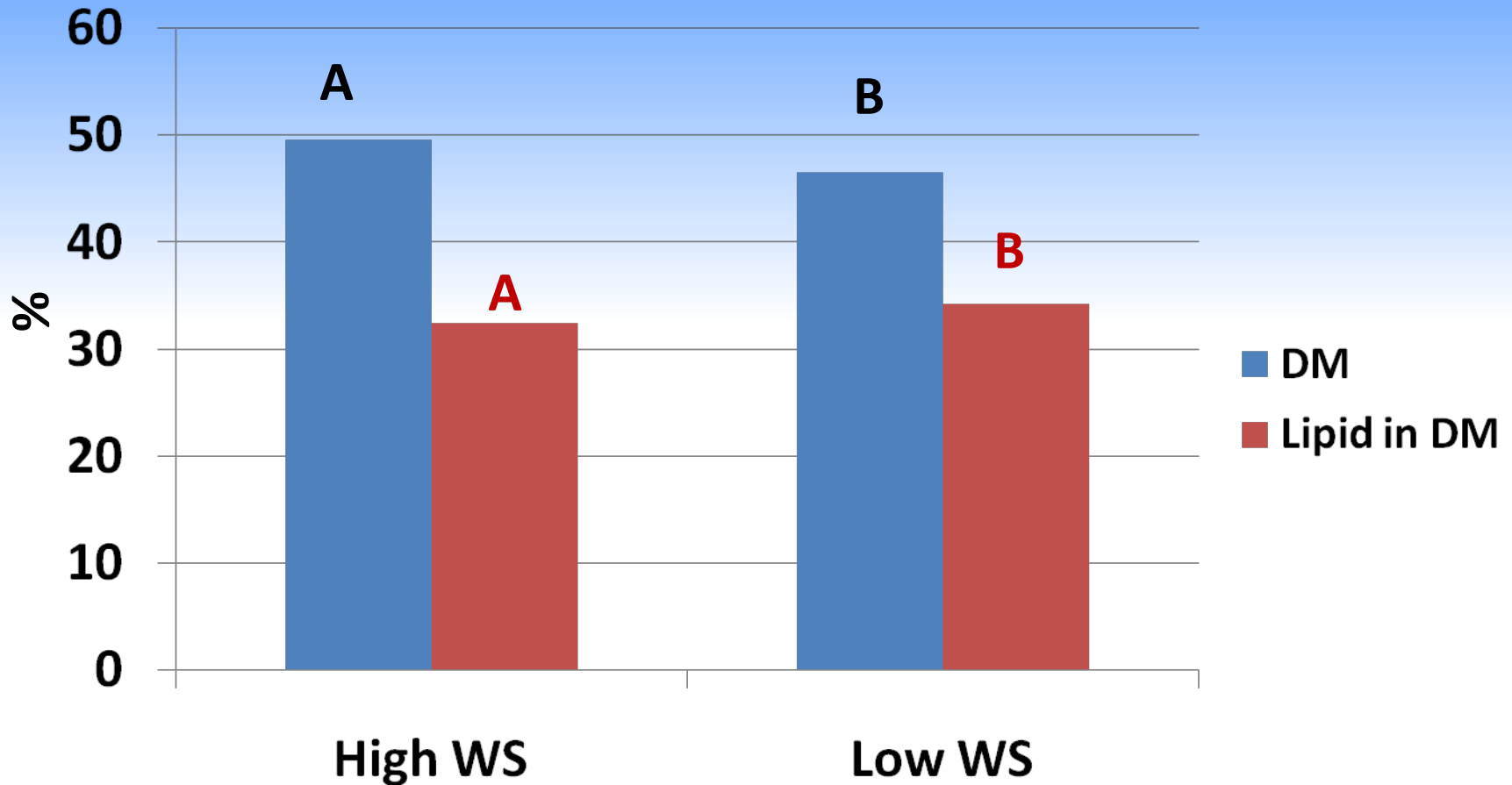
$$\text{Oil absorption capacity} = 100 * (W2 - W1) / W1$$

Water Stability of Pellets

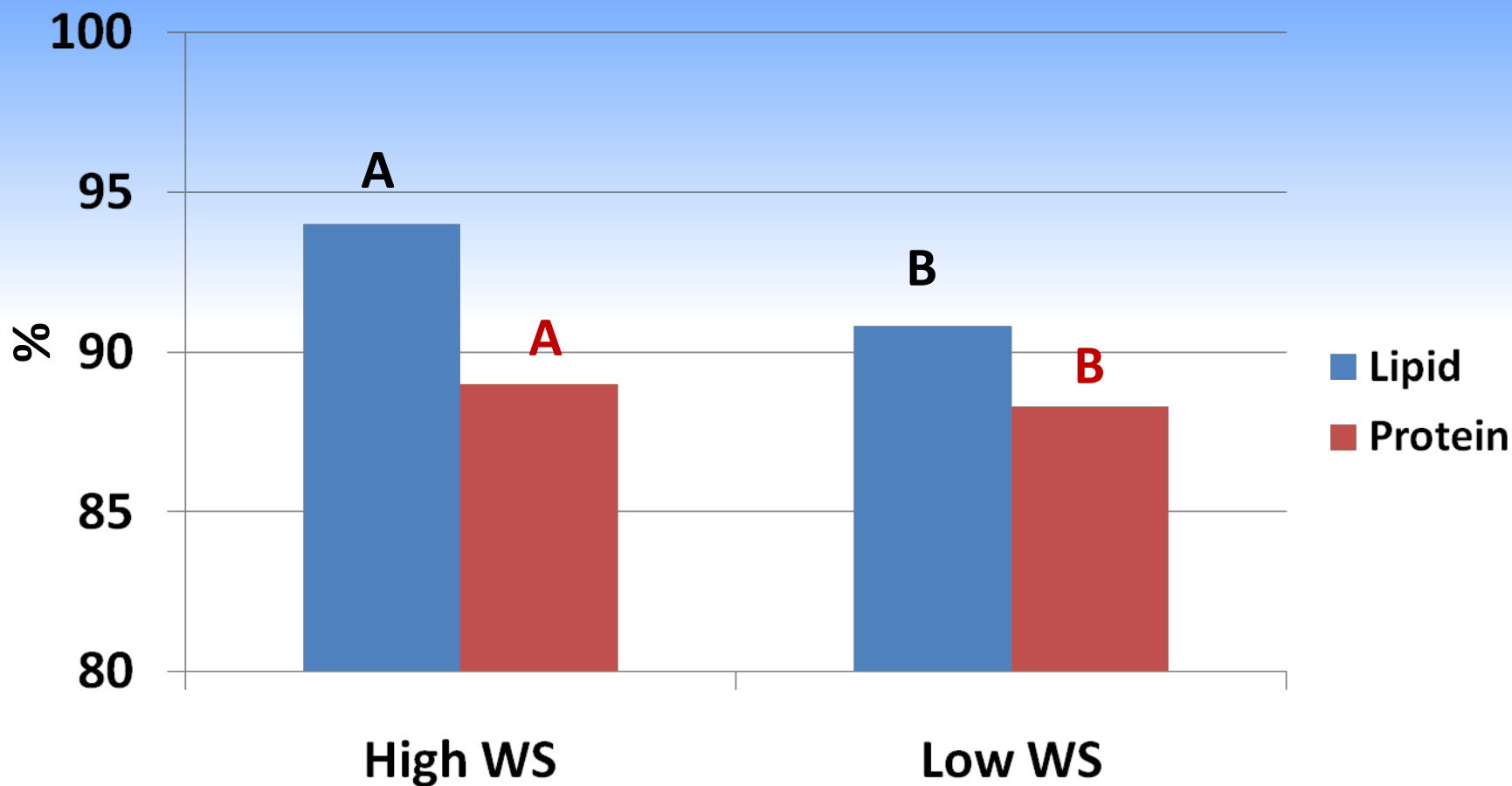


$$\text{Water stability}\% = \frac{100 * \text{dry matter of retention pellets (g)}}{\text{dry weight of original pellets (g)}}$$

Pellet Water Stability Affects the Dry Matter and Lipid levels of Stomach Contents



Water Stability of Pellets Affects Apparent Digestibility of a Diet



Summary

- **Different ingredients can significantly affect both physical and chemical quality of feeds !**
- **Different ingredient may require different feed processing method !**
- **Processing method is important for quality of an ingredient as well as a feed !**

Evaluation of an Ingredient

Attractability/Palatability Test



Measurement of Food Intake



Evaluation of an Ingredient

Digestibility

In vitro

In vivo

- Manual stripping; Rectal suction
- Siphoning; Settlement
- Dissection



Evaluation of an Ingredient

Growth Trial



Green water system

Clean water system

Integrated Researches

Nutritional studies

Biochemistry

Feed processing technology

Substitution of Fishmeal Protein by an Algae Meal in Feed for Pacific Threadfin (Moi)

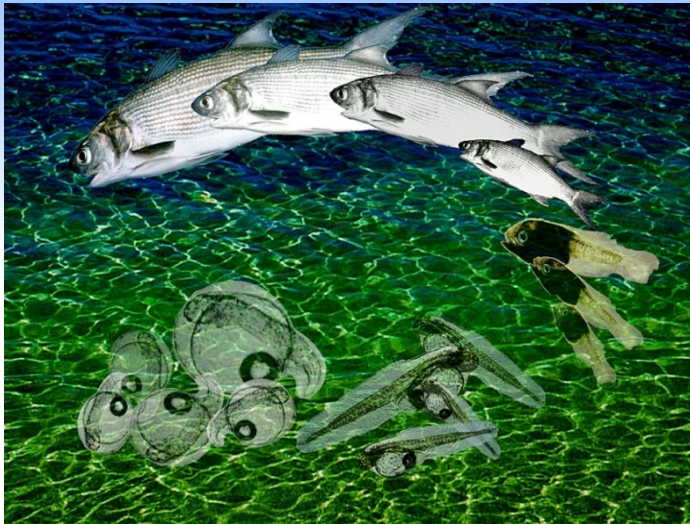


Fig. 8 Fresh Moi being prepared for steaming.

Commercial Feed

48-50% protein
14% lipid



OI Feed

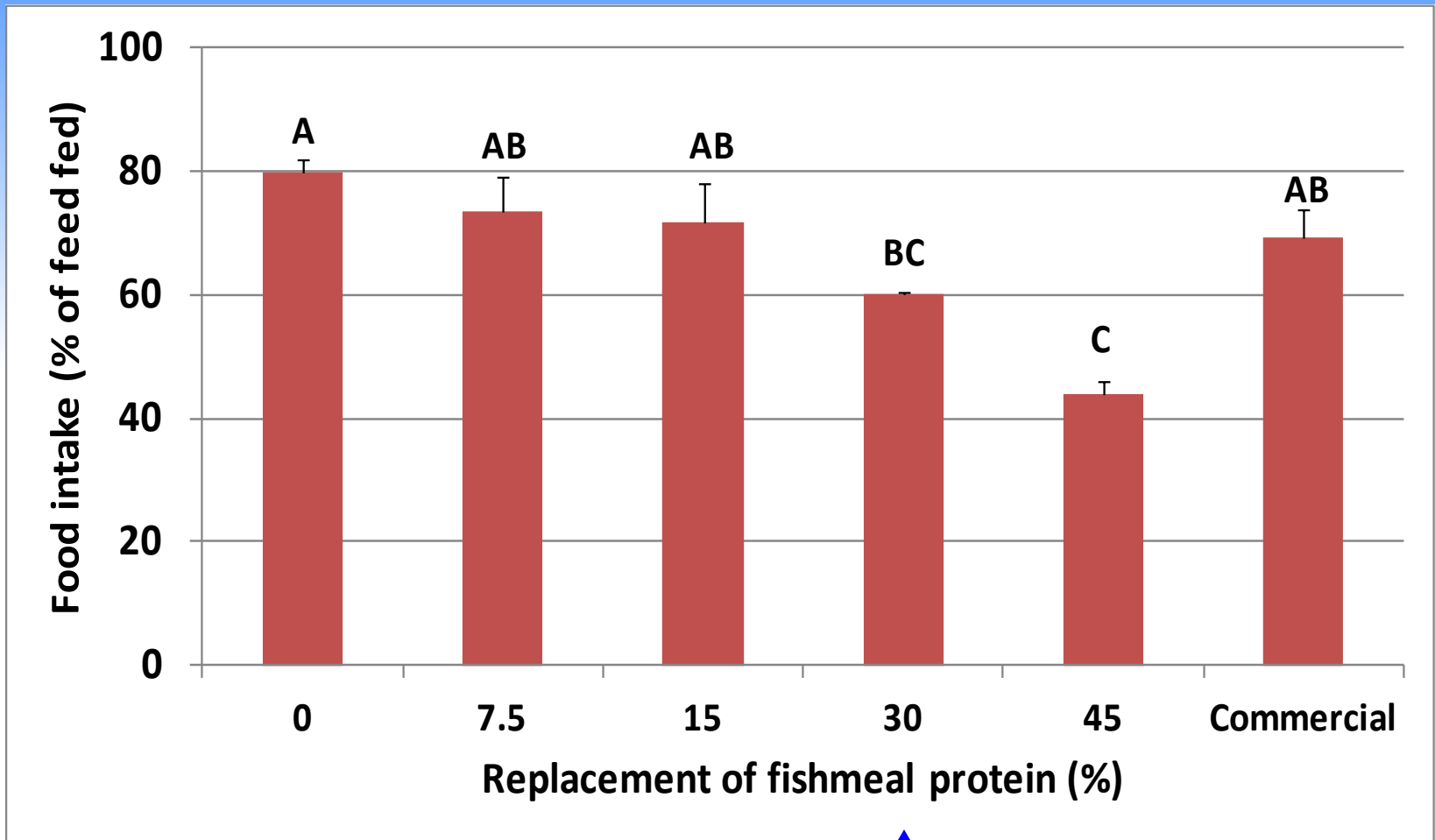
33-40% protein
10-12% lipid

Formulation of Test Diets for Juvenile Moi

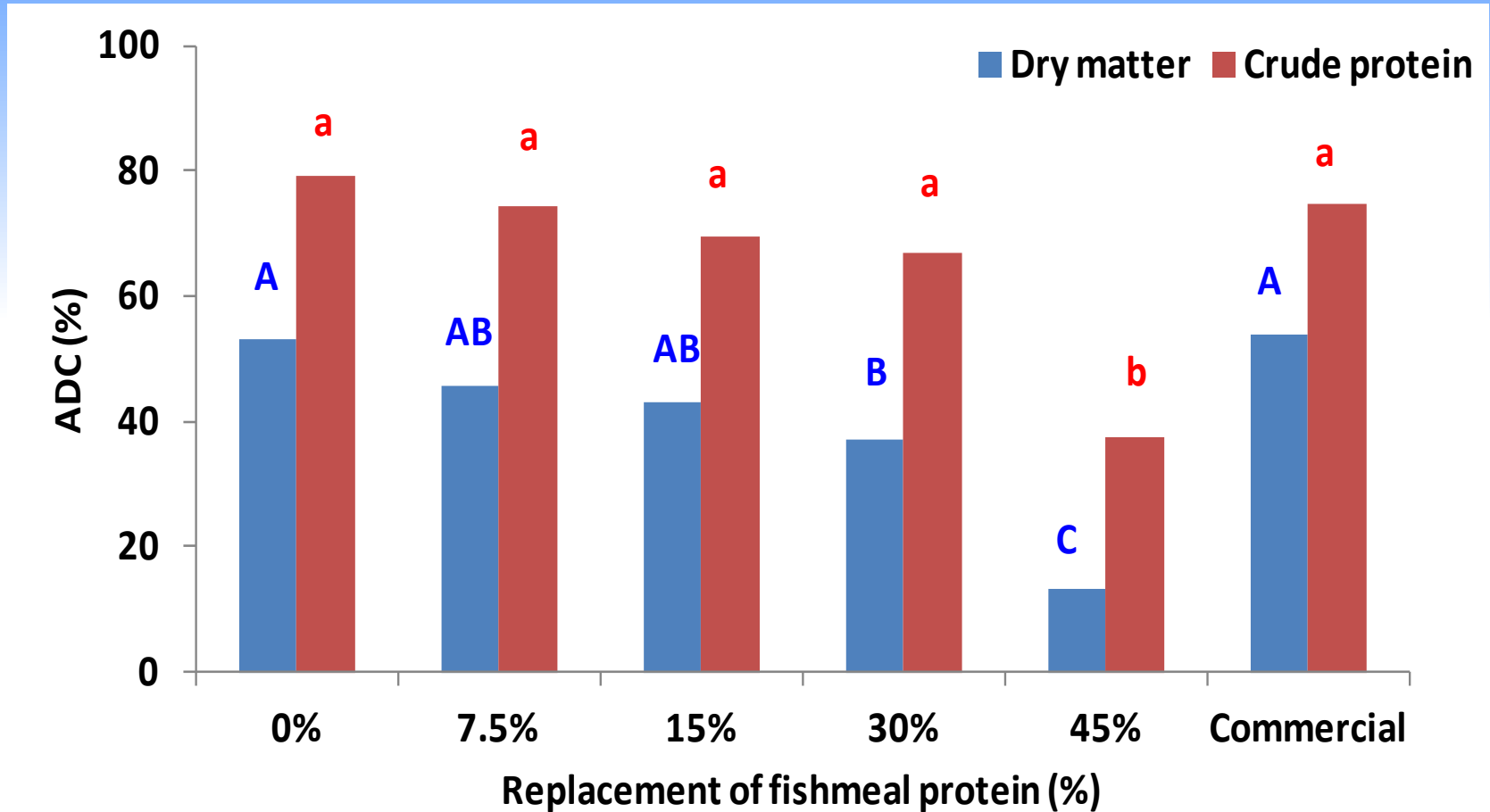
Ingredients	Replacement of fishmeal protein (%)				
	0%	7.5%	15%	30%	45%
Pollock meal	30	27.8	25.6	21.2	16.8
Soybean meal	20	20	20	20	20
Algae meal (29.5% CP)	0	5	10	20	30
Others	50	52.8	55.6	61.2	66.8

Crude protein: 37% Crude lipid: 13%: Gross energy: 19.3 kJ/kg

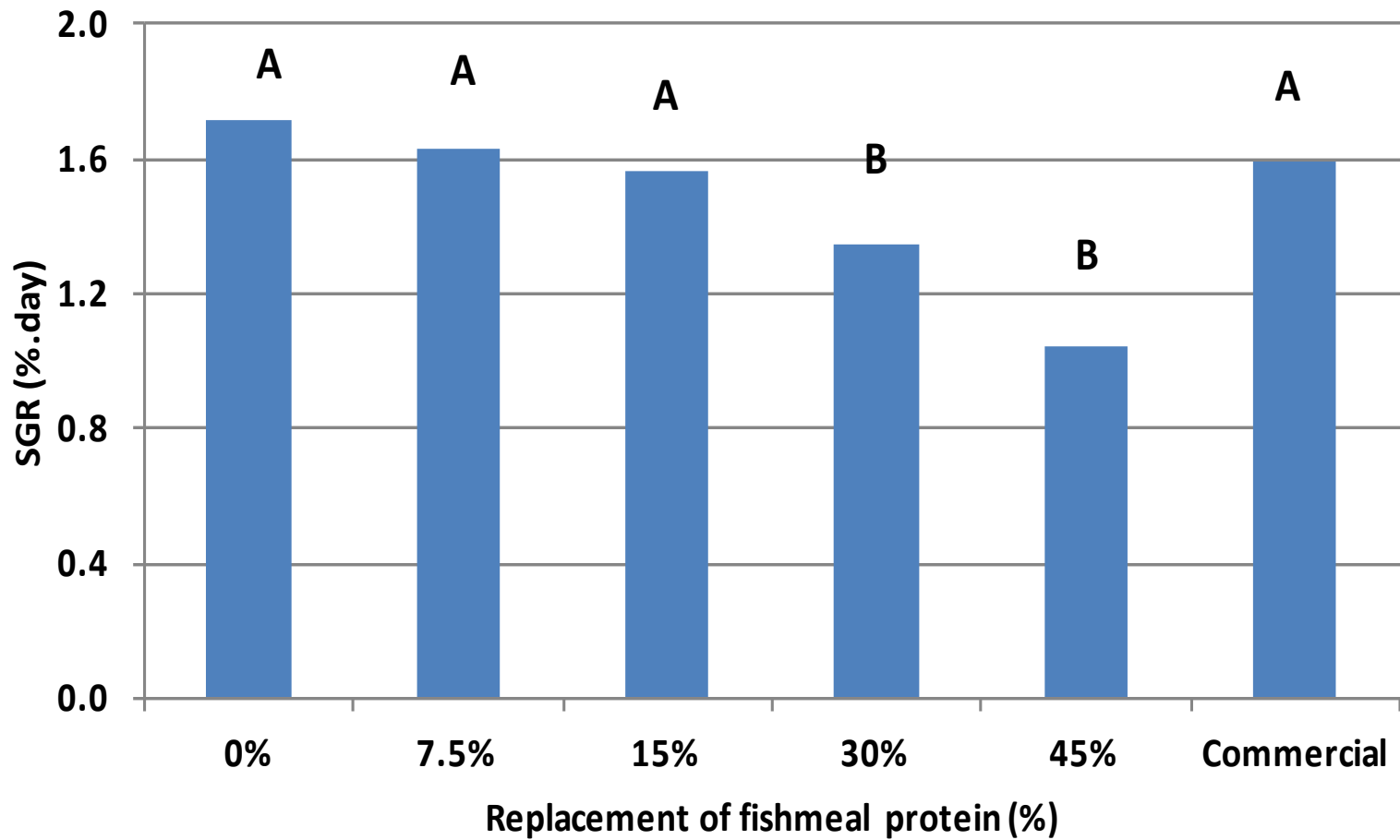
Effect of Algae Meal on Palatability of Fish



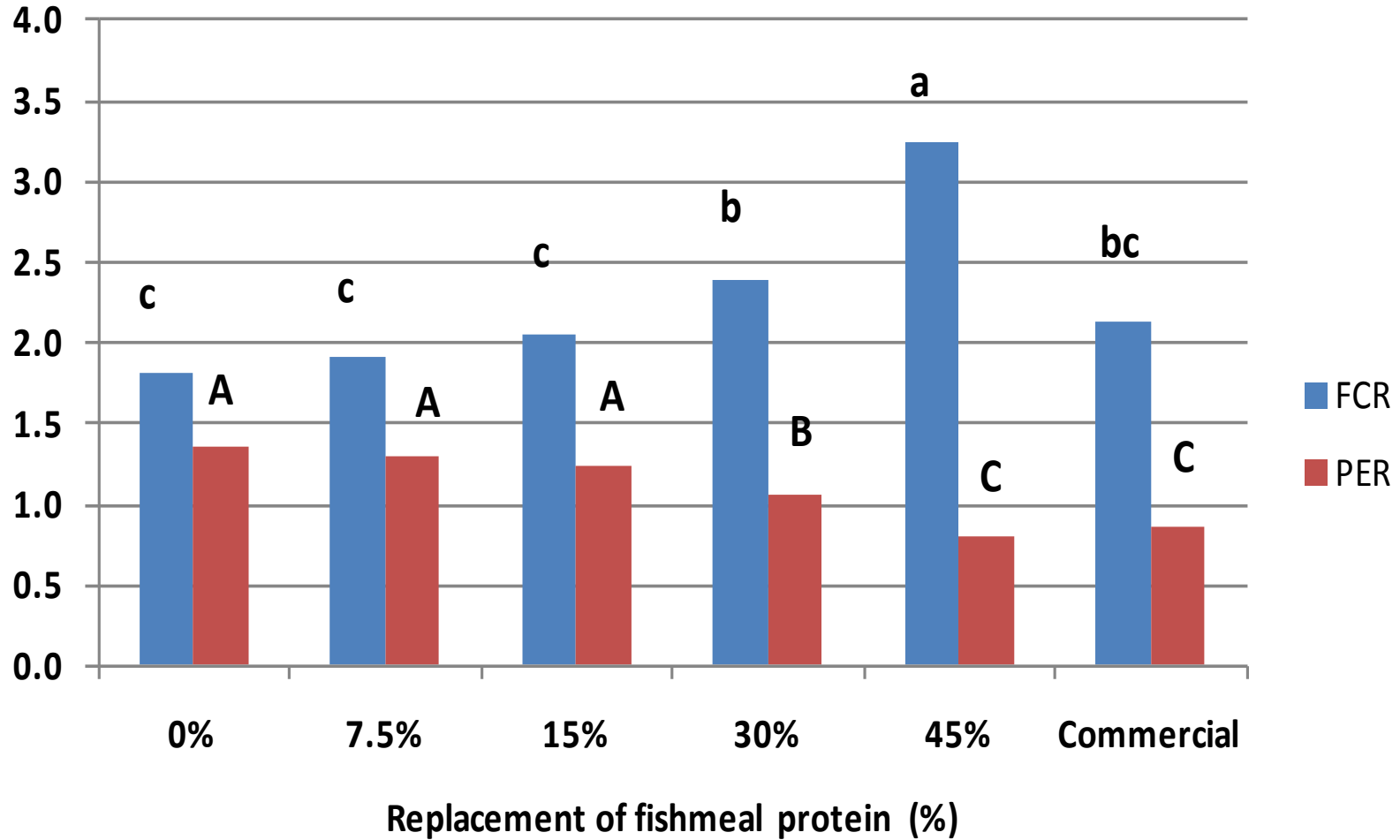
Effect of Algae Meal on Digestibility of Dietary Nutrients for Fish



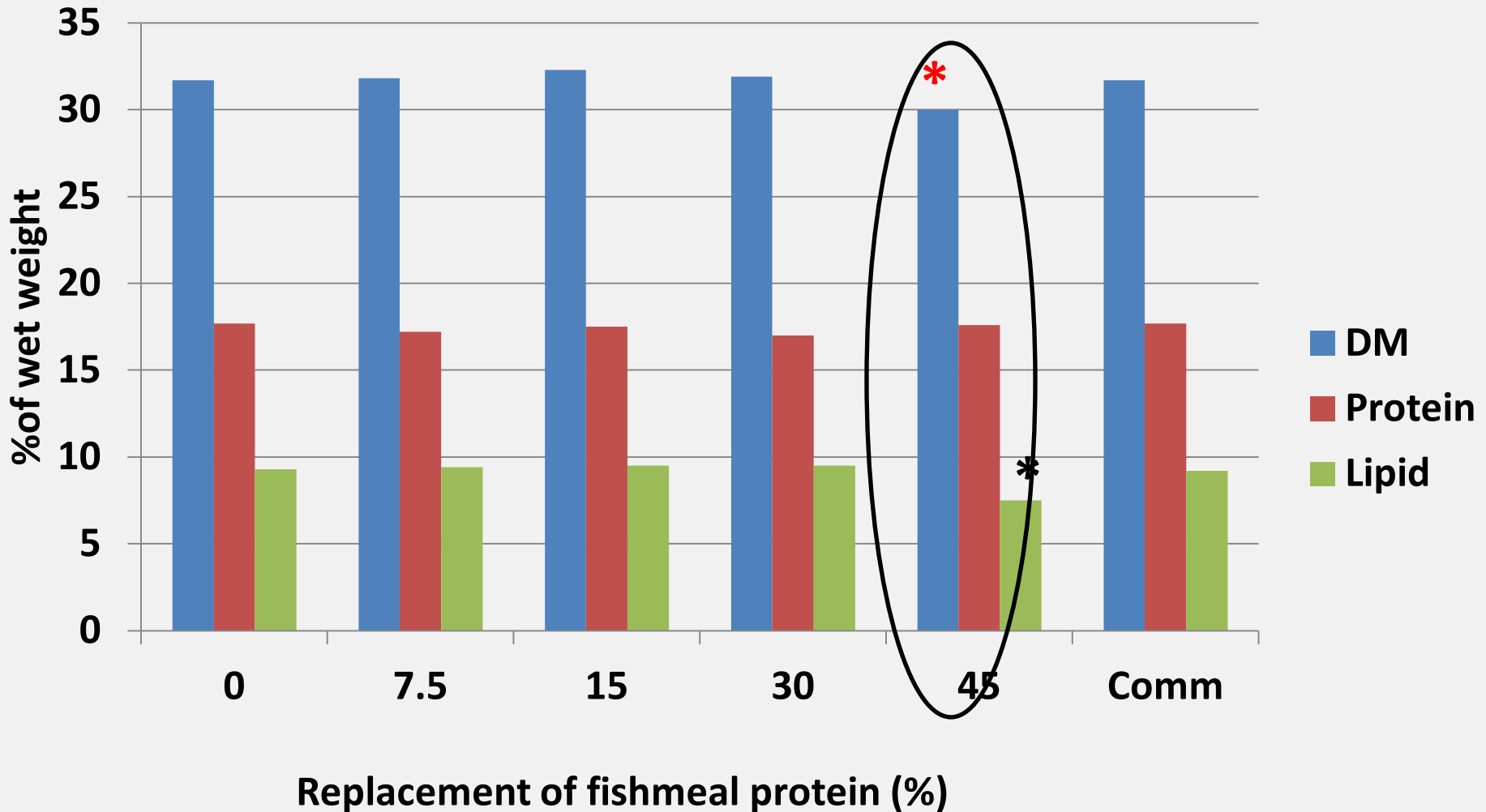
Effect of Algae Meal on Growth of Fish



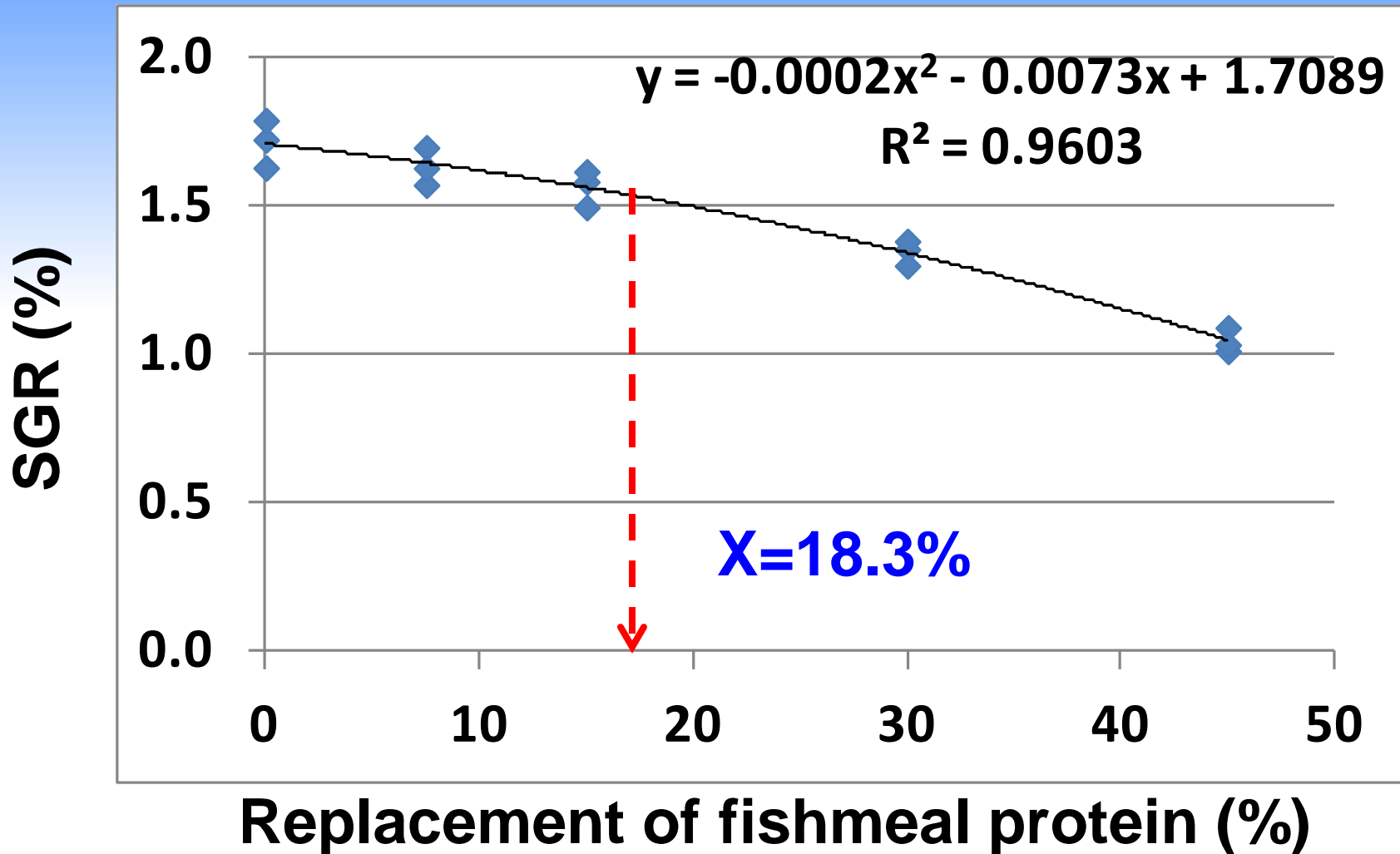
Effect of Algae Meal on Feed Utilization



Effect of Different Diets on Nutritional Composition of Fish Fillet



Optimal Replacement Level of Fish Meal Protein by the Algae Meal



Amino Acids Contents of Test Diets and Ingredients

Amino acids	Commercial	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Fishmeal	Algae
<u>Dispensable AA</u>		g/100 g diet as fed						
Taurine	0.46	0.50	0.46	0.41	0.33	0.28	2.74	0.02
<u>Indispensible AA</u>								
Methionine	1.08	0.81	0.79	0.77	0.71	0.70	4.94	0.34
Phenylalanine	2.37	1.67	1.65	1.56	1.52	1.46	2.09	1.66

Summary

- **The algae meal can replace 18% fishmeal protein based on performance of fish;**
- **The substitution level can be up to 30% based on feed utilization and nutritional composition of fish;**
- **Deficiency of amino acids in the algae meal may be one of the reasons for the poor performance of fish fed the high algae diet.**

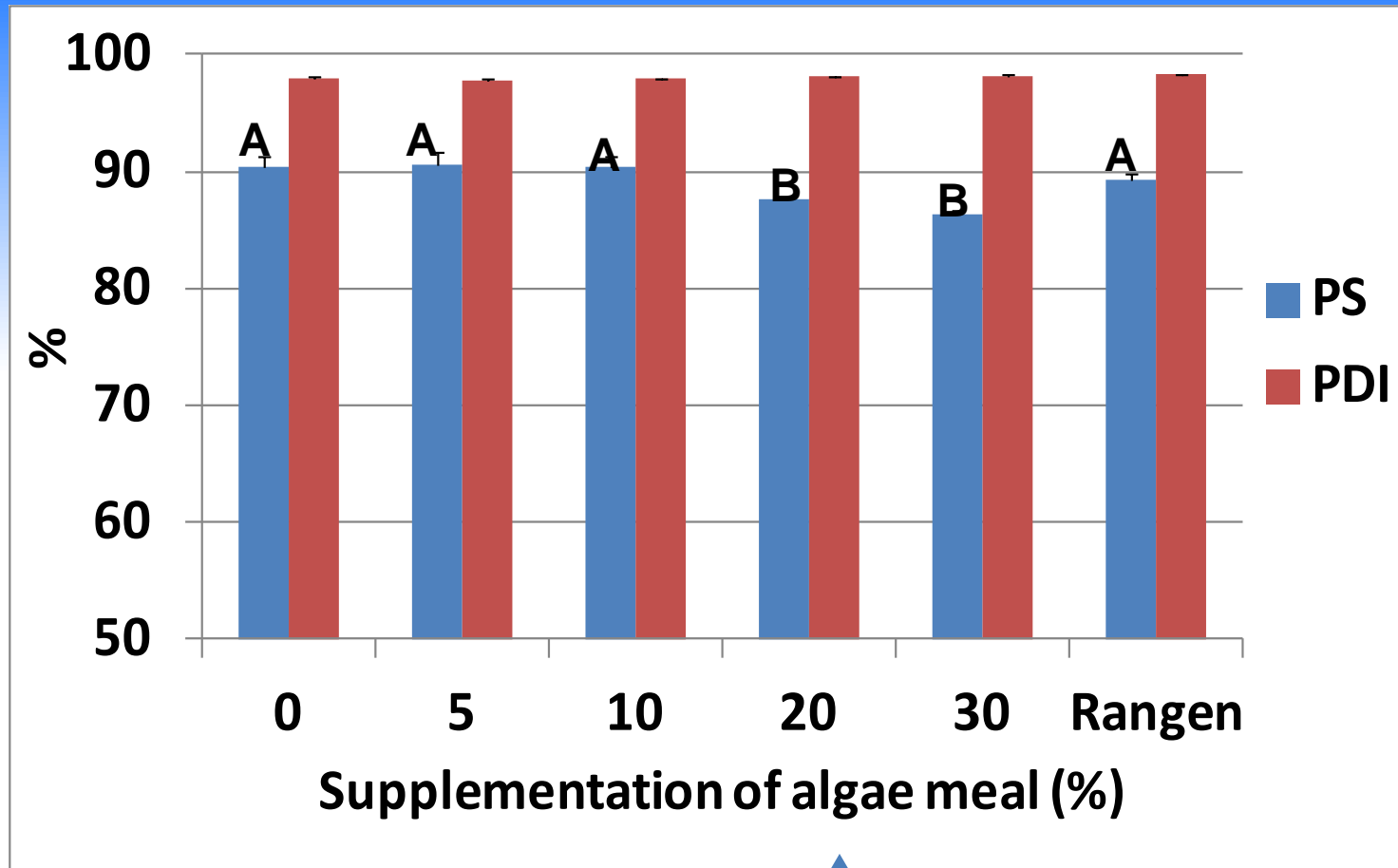
Substitution of Fishmeal Protein by an Algae Meal in Shrimp Feeds

Formulation of Test Diets for Shrimp

Ingredients	Replacement of fish meal protein (%)				
	0%	16%	32%	64%	100%
Menhaden meal	15	12.7	10.4	5.8	0
Soybean meal	25	25	25	25	25
Algae meal (29.5% CP)	0	5	10	20	30
Others	60	57.3	54.9	49.2	45

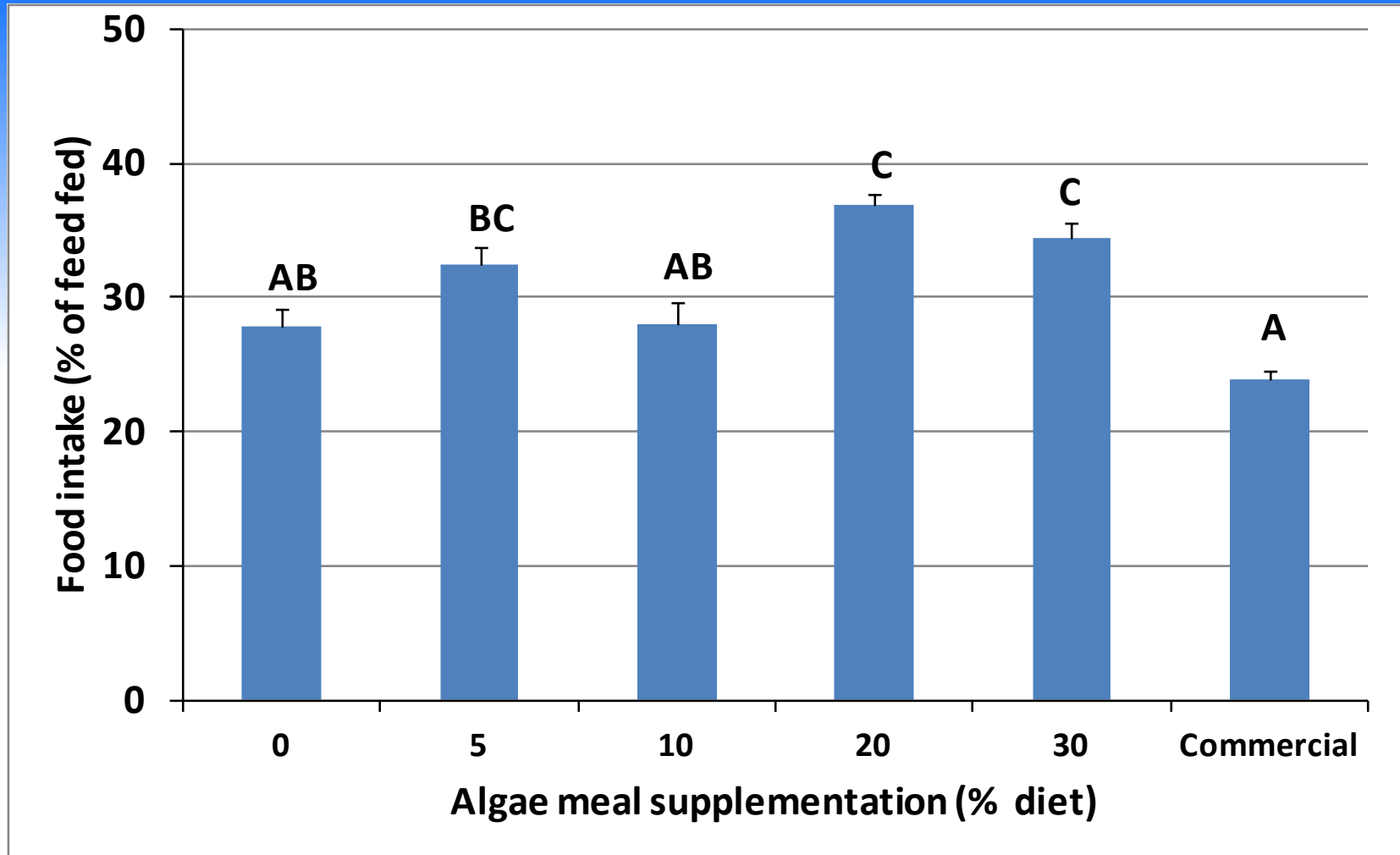
Crude protein: 33%; Crude lipid: 9%

Water Stability and Durability of Pellets

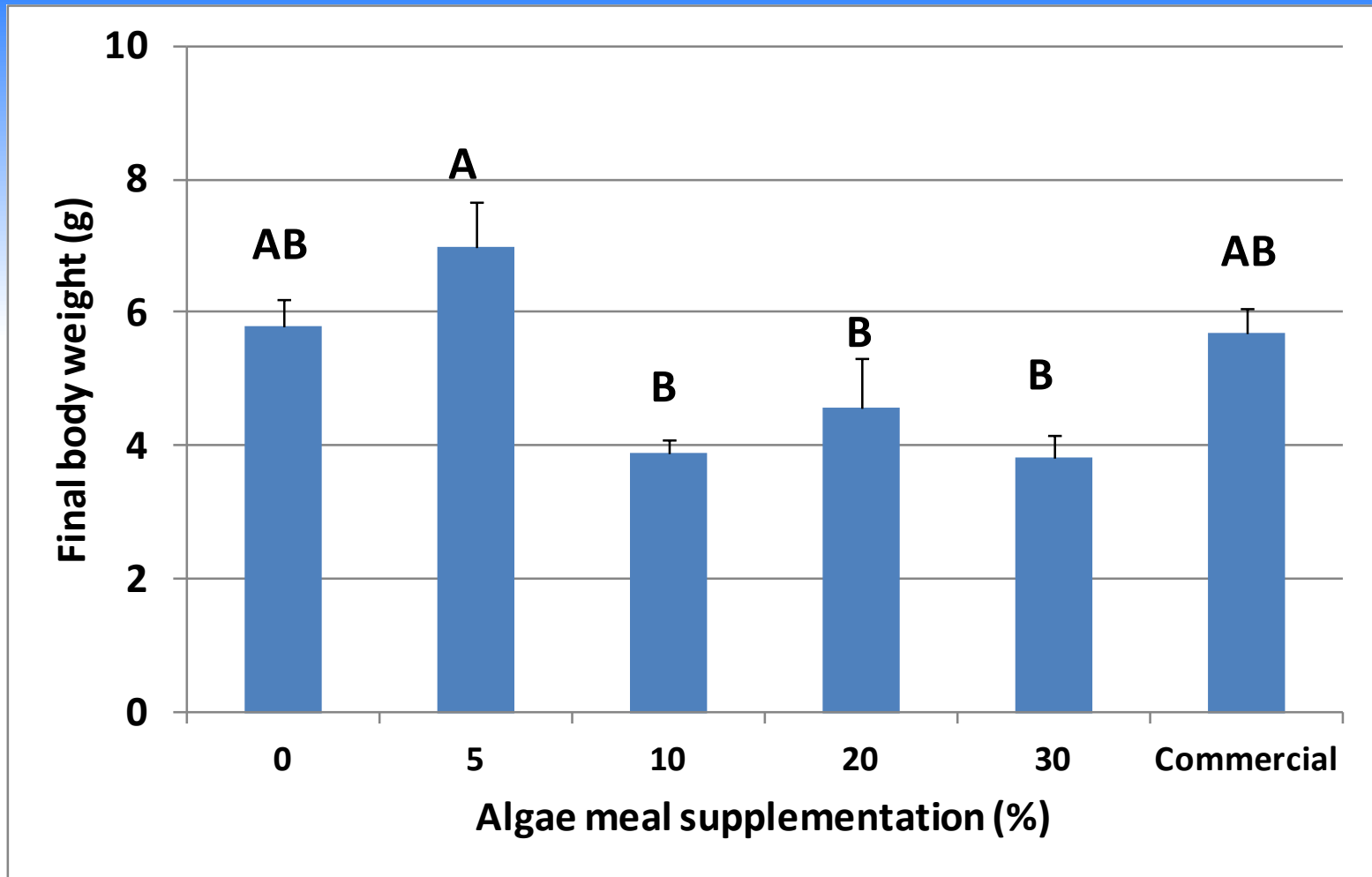


Replaces 64% fishmeal protein

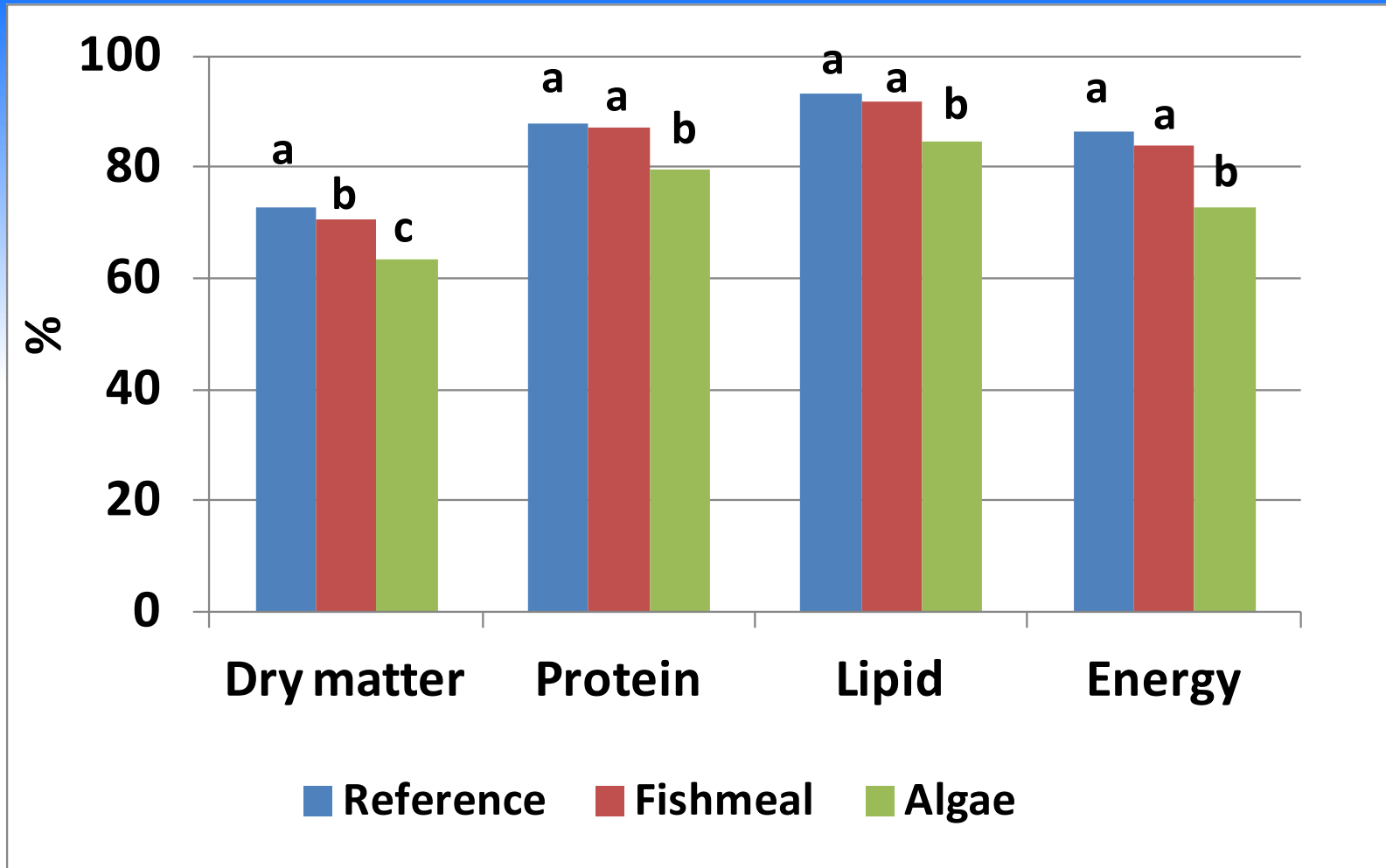
Algae Meal Enhances Palatability of Shrimp



Growth of Shrimp Fed Diets with Different Levels of Algae Meal



Digestibility of Dietary Nutrients in Shrimp



Test diet for digestibility: 30% ingredient +70% reference diet

Summary

Algae meal tested

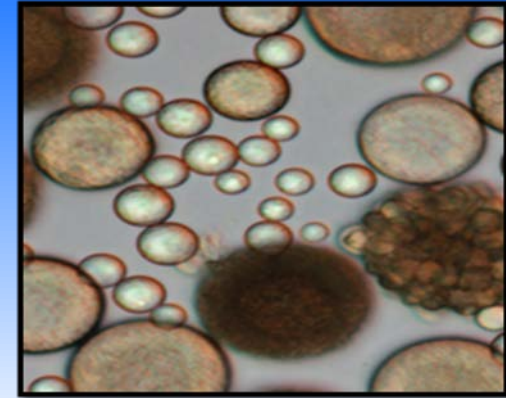
- Enhances shrimp palatability;
- Does not affect pellet durability but decreases water stability of pellet;
- Tends to decrease growth of shrimp;
- Has lower digestibility than fish meal.



Sources of Lipid/Long Chain PUFA



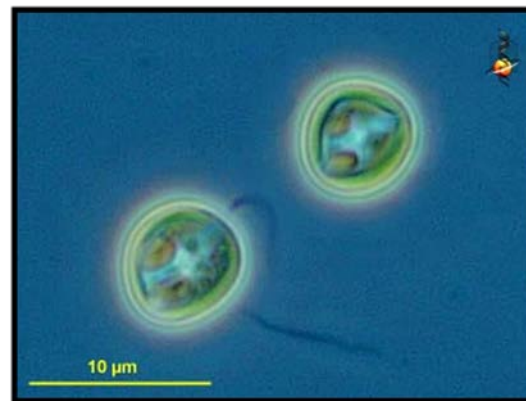
Dinoflagellate alga (*Cryptothecodinium Cohnii*)



Thraustochytrids (*schizochytrium*)

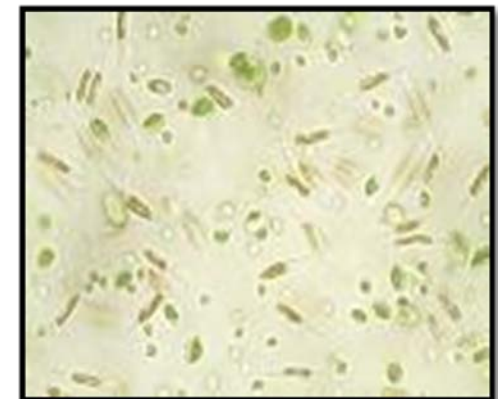


Isochrysis galbana



Pavlova lutheru

<http://phytoonline.mdamirpp.net/>



Nannochloropsis oculata

Fatty Acids Profiles of Different Ingredients

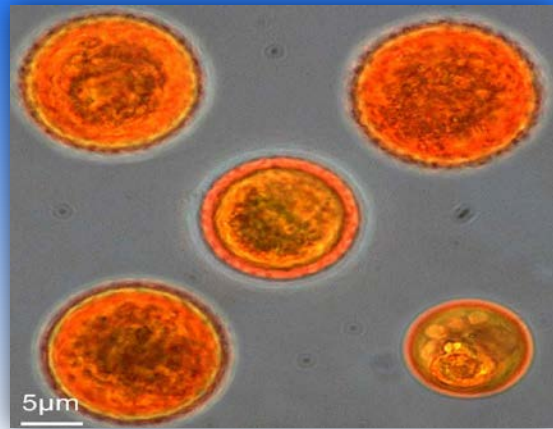
(% of Total Fatty Acids)

Ingredients	18:2n-6	18:3n-3	20:4n-6	20:5n-3	22:6n-3	Chol.
Menhaden oil	1.3	0.3	0.2	11	9.1	0.52
Cod liver oil	1.4	0.6	1.6	11.2	12.6	0.57
Tallow oil	3.1	0.6	---	---	---	0.1
Soybean oil	51	6.8	---	---	---	---
Corn oil	58	0.7	---	---	---	---
Chaetoceros sp	1	0.4	3	16.7	0.8	
Pavlova lutheri	2.1	2.1	0.5	28.3	15.5	
Isochrysis galbana	8.6	4.5	---	0.9	19.4	
Cryptomonas sp	0.6	25.1	0.2	12	6.6	
Rhodomonas sp	1.9	25.2	---	8.7	4.6	
Schizochytrium sp	0.7	0.11	2.9	0.6	31.4	

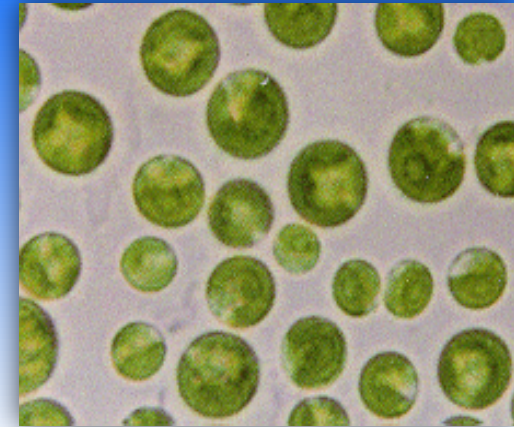
Chol, cholesterol, % of diet

(NRC, 1993, 2011; Reitan et al. 1997 and Renaud et al., 1999. Aquaculture)

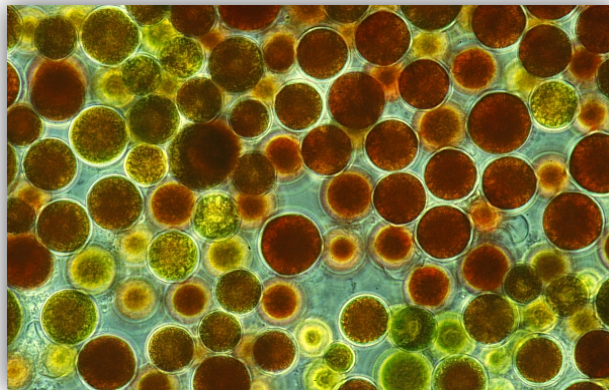
Nutritional Pigments



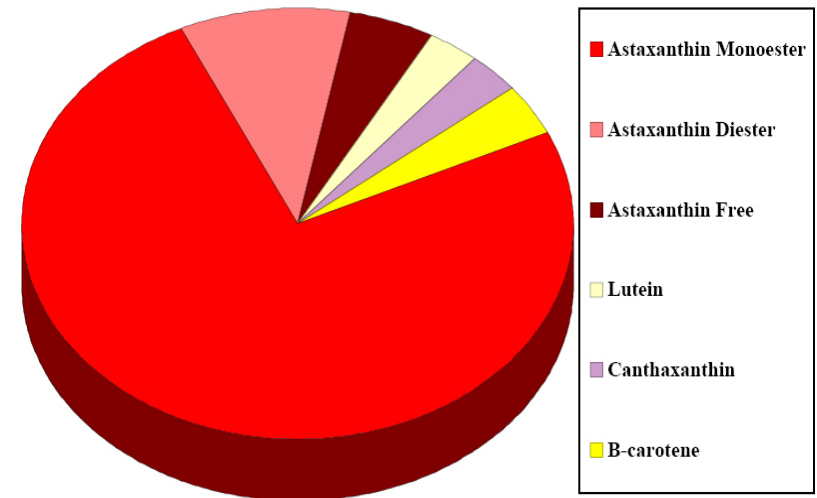
Dunaliella Salina produces β -carotene



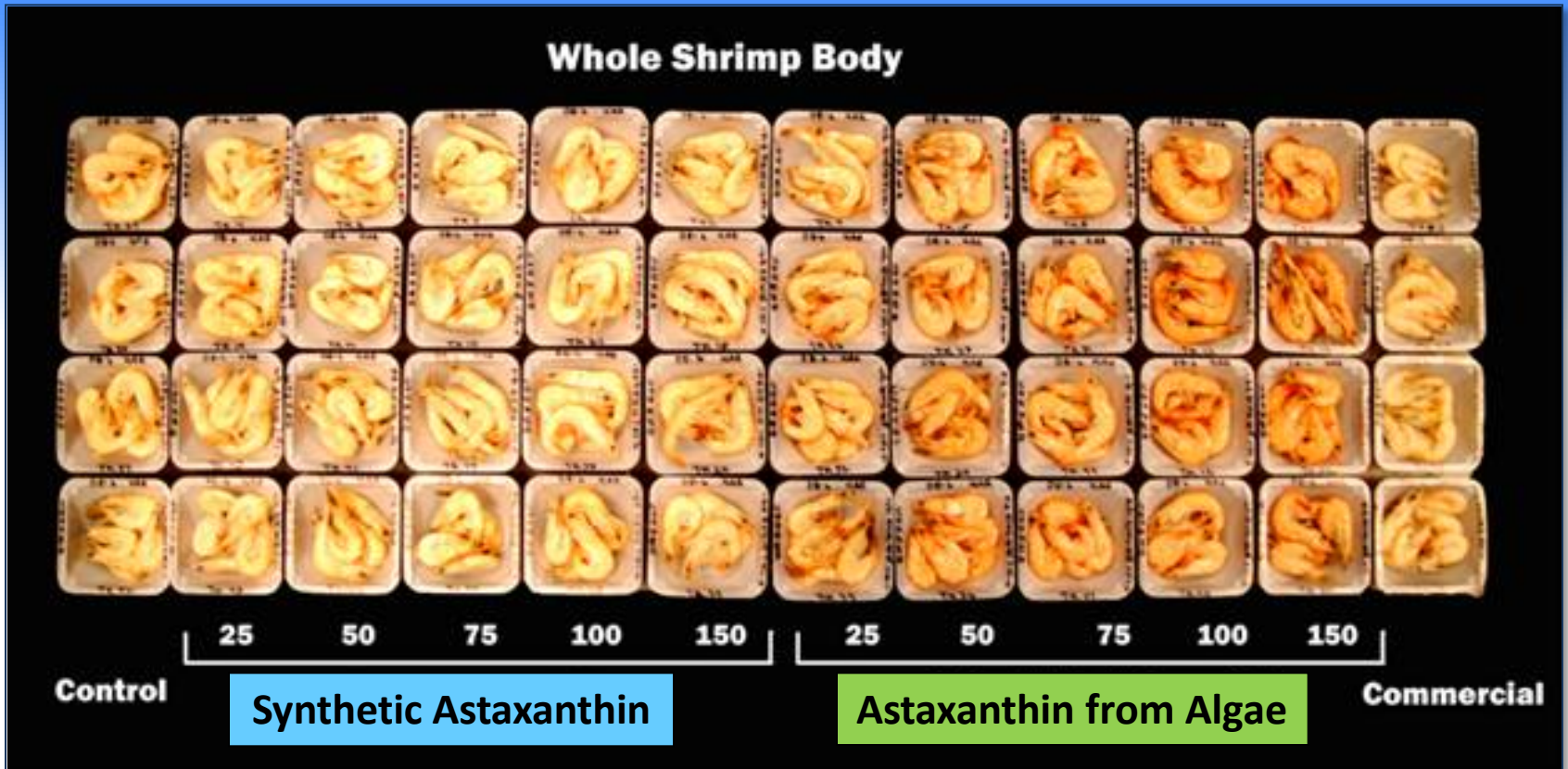
Chlorella produces lutein and astaxanthin



Haematococcus produces mixed carotenoids



Astaxanthin Improves Pigmentation in Shrimp

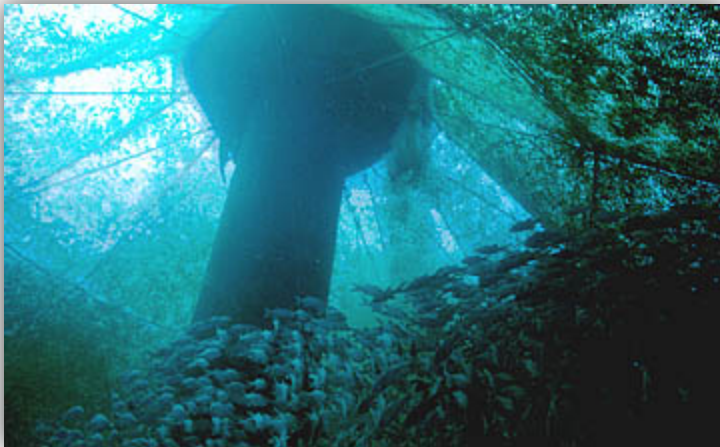


Concerns

- **Nutrient levels and balance**
- **Anti-nutritional factors or contaminants**
- **Availability of nutrient (digestibility, palatability)**
- **Processing methods for ingredients and feeds**
- **Quality of end product (flavor, texture and nutritional values)**
- **Production and cost of a by-product**

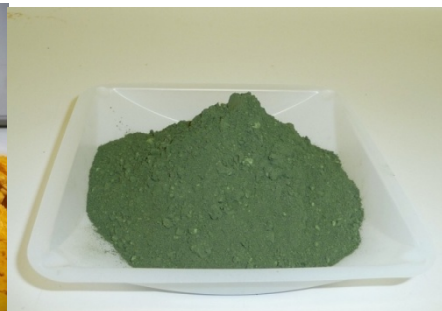
Future Research

- **More research to update nutrient requirements and thus formulation for different farmed fish**
 - **Culture system/condition**
 - **New species or family of animal**



Future Research

- **Multidisciplinary collaborative researches**
 - ingredient selection
 - selective breeding for optimal family
 - processing technology development



Future Research

- **Standard research diet development**
 - Ingredients**
 - Formulation**
 - Feed processing protocol**
 - Evaluation methods**



Acknowledgements

- Funding by Agricultural Research Service, United States Department of Agriculture

