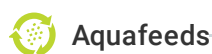




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Cholesterol: Indispensable but not irreplaceable in shrimp feeds

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Specialty concentrate enhances digestion and absorption of endogenous cholesterol in shrimp feed ingredients

Penaeid shrimp are unable to bio-synthesize cholesterol, which is widely accepted as an essential nutrient for these crustaceans. It is a key constituent of cell membranes and a precursor for steroid and moulting hormones.

Increased demand for cholesterol, combined with its relatively high cost and variable supply present a challenge for the shrimp feed industry. However, progress has been made in the development of cost-effective alternatives to purified cholesterol.

In laboratory trials with *Penaeus monodon*, the authors formulated a specialty concentrate that enhances digestion and absorption of endogenous cholesterol in shrimp feed ingredients and reduces the need to supplement feed with purified cholesterol.



Feed trials at the Brackishwater Aquaculture Development Center in Indonesia examined cholesterol's impact on shrimp feed.

Sources and availability

Meals and oils from marine invertebrates like squid, shrimp, clams, crabs and mussels are good sources of cholesterol. However, fishmeal is often the major cholesterol source in practical feed formulations for shrimp.




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Feed-grade cholesterol is also derived from wool grease, a byproduct collected in the washing of sheep wool. The cost and availability of cholesterol fluctuated during the past decade, due to large variations in the market for wool grease (lanolin). Diminishing sheep stocks and increasing competition for lanolin derivatives (e.g., steroids) may become serious concerns for formulators of shrimp feeds in the future.

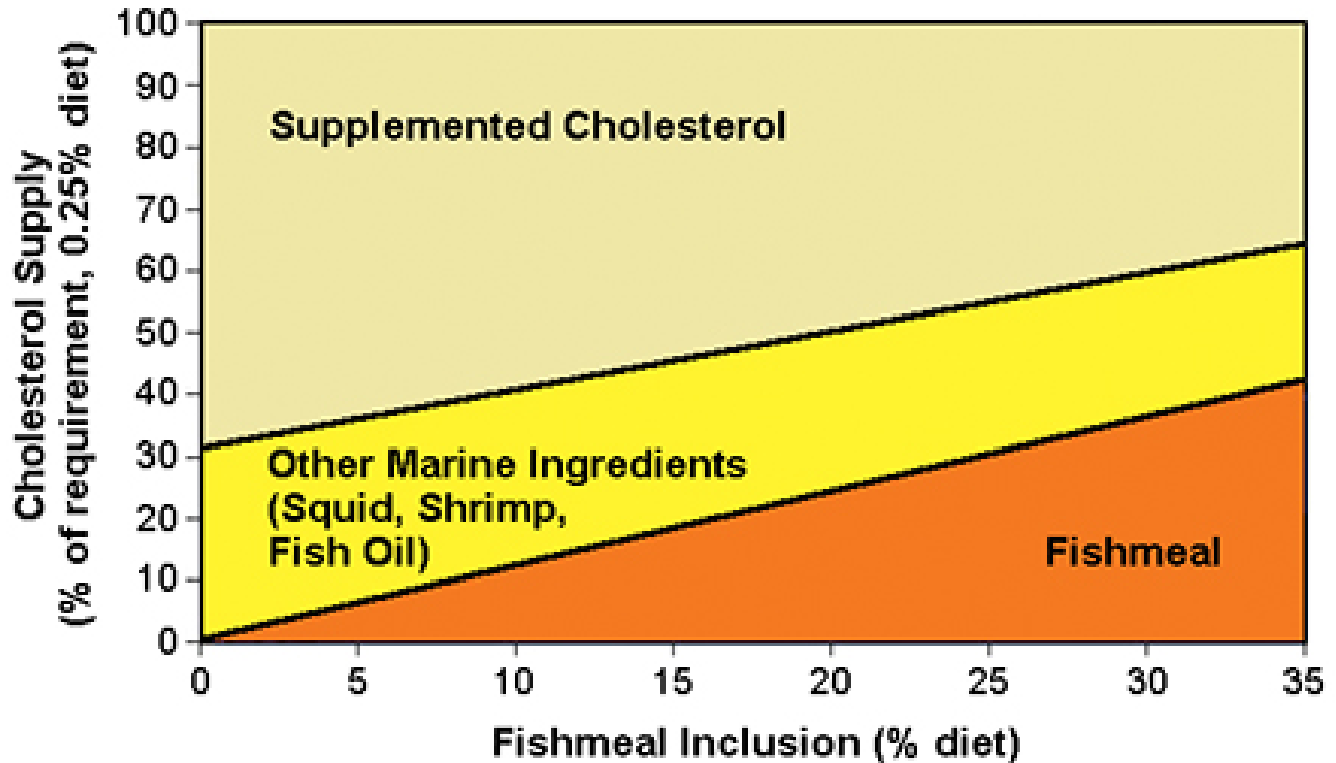


Fig. 1: Effect of replacement of fishmeal by plant proteins on cholesterol sourcing in typical shrimp feed formulated to contain 0.25 percent cholesterol. Graph shows the percentage of total dietary cholesterol provided by fishmeal, other marine ingredients, and purified cholesterol as a function of fishmeal inclusion. Calculation is based on average levels of cholesterol in marine raw materials and assumes an increase of other marine ingredients with decreasing fishmeal inclusion for feed attractivity.

Tacon and Forster (2001) foresaw global aquafeed requirements for fishmeal to grow from 15 million metric tons in 2000 to 27 million MT by 2010. This will result in an increased demand for – and possible shortage of – marine raw materials.

Although animal byproducts offer interesting nutritional alternatives as protein sources for penaeid shrimp, their use is not accepted by European consumers and legislators. Therefore, the need for additional supplementation of cholesterol in shrimp feeds is likely to increase, due to the rising pressure on fishmeal as a protein source in shrimp feeds.

Assuming a minimum dietary level of 0.25 percent cholesterol, fishmeal at 35 percent inclusion in shrimp diets can provide around 50 percent of the requirement (Fig. 1). The addition of typical levels of shrimp and squid meal usually supplies another 20 to 25 percent. Thus, supplementation of purified

cholesterol at 0.10 to 0.15 percent of the diet is needed to fully cover the requirement, and represents a significant cost in shrimp formulations.

Feed preparation and ingredients

For the trials, practical diets were prepared by lab scale pelletizing (Table 1). Diets were formulated to satisfy documented nutritional requirements of *P. monodon*, except for cholesterol (Akiyama et al., 1992).

Coutteau, Feed formulation for experimental diets, Table 1

Ingredient	CHOL-	CHOL+	CHOL-repl
Standard fishmeal (70% CP/10 CF)	33.7	33.7	33.7
Shrimp head meal	4.0	4.0	
Defatted soybean flour	26.4	26.4	26.4
Wheat middlings	8.5	8.5	8.5
Pregelatinized wheat starch	20.0	20.0	20.0
Soybean oil	1.0	1.0	1.0
Sodium alginate	2.0	2.0	2.0
Shrimp premix*	3.0	3.0	3.0
Cholesterol (85% purity)		0.18	
Cholesterol-replacing premix**			
Wheat middlings (filler)	1.5	1.32	
Total	100	100	100
Proximate composition (% diet)			
Crude protein	40.3	39.3	39.5
Crude fat after hydrolysis	7.3	7.4	7.2
Crude ash	8.8	9.1	9.2
Crude fiber	2.9	3.1	3.0
Moisture	7.1	6.9	7.2

* Nutritional concentrate providing vitamins, minerals, trace elements, and essential amino acids.

** Cholesterol-replacing concentrate providing a blend of nutrients that enhance absorption and utilization of dietary cholesterol.

Table 1. Feed formulation for experimental diets in trial 1 (% as fed).

Three feed formulations were compared:

- A control diet (CHOL-) with a background concentration of 0.10 percent cholesterol, but no cholesterol supplementation.
- A positive control diet (CHOL+) supplemented with 0.15 percent cholesterol to a total level of 0.25 percent cholesterol.
- A test diet (CHOL-repl) supplemented with cholesterol replacement.

To verify initial findings, a second batch of feeds was prepared following the same guidelines, but using different batches of raw materials. This resulted in slightly different proportions of fishmeal (35 percent) and defatted soybean meal (25 percent), and a higher background of cholesterol in the feeds. CHOL-, CHOL+, and CHOL-repl contained 0.15, 0.30, and 0.15 percent cholesterol, respectively. Dietary cholesterol analysis was performed using GL chromatography.

Culture trials

Two culture trials were conducted at the Brackishwater Aquaculture Development Center in Indonesia.

Tanks and water

The experimental setup consisted of nine cylindrical, 1-metric tons (MT), flat-bottom tanks. Seawater was prepared by pumping through a 1- μ m GAF filter bag, and recirculated over a 1.6-MT coral biofilter.

Salinity was adjusted to 25 ppt by the addition of underground freshwater, and disinfection with 3 milligrams per liter hypo-chlorite powder for 24 hours. Mean water renewal over the whole culture period was 28 percent per day.

Shrimp of 0.3 to 0.4 grams were initially stocked at 20 shrimp per tank and acclimated for one week prior to the start of the trial. Water temperature (27.5 ± 0.7 degrees-C), salinity, and other quality parameters remained within acceptable limits during tests.

Survival, growth, FCR

Average survival during the first trial was over 88 percent (Table 2). Overall shrimp growth up to 1.4 grams per week toward the end of the trial was excellent under the conditions of clear water culture in small tanks. Feed-conversion ratios of 3.1 to 3.9:1 were adequate for shrimp feeding in clear water tanks without natural food organisms.

Coutteau, Results after growing *P. monodon* 70 days on experimental feeds, Table 2

Parameter	CHOL-	CHOL+	CHOL-repl
Survival (%)	88.3	90.0	91.7
Final individual weight (g)	5.86 ^b	7.78 ^a	7.48 ^a
Growth rate (%/day)	3.15 ^b	3.57 ^a	3.48 ^a
Final total biomass (g)	103.2 ^b	137.3 ^a	133.8 ^a

Feed-conversion ratio	3.9 ^b	3.1 ^a	3.4 ^a
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Table 2. Results after growing *P. monodon* 70 days on experimental feeds (Trial 1). Different letters denote significant differences.

Cholesterol supplementation

Fig. 2: Shrimp biomass per tank after 70 days for Trial 1, and 56 days for Trial 2. Data represent average of group weights from triplicate tanks.

Growth and feed conversion were significantly improved by the supplementation of 0.15 percent cholesterol. After 70 days of culture, harvested biomass was 33 percent higher for the shrimp fed the diet supplemented with cholesterol (Fig. 2). This was not due to density differences, but solely to dietary components.

Cholesterol replacement

The cholesterol-replacing concentrate was as effective as purified cholesterol in improving growth and feed conversion for the negative control diet. After 70 days of culture, harvested biomass was 30 percent higher in the CHOL-repl treatment than the negative control.

Overall shrimp survival was lower in trial 2, with treatment averages ranging 77 to 85 percent. However, the trends in growth response observed in this independent trial – run with different shrimp and different batches of raw materials for feed preparation – generally matched those of the first trial.

Conclusion

These studies confirmed that the requirement for dietary cholesterol in *P. monodon* exceeds the endogenous background level typically provided by ingredients of marine origin in practical diets. The supplementation of 0.15 percent purified cholesterol or 1.5 percent of a cholesterol-replacing concentrate to a practical diet with a cholesterol level of 0.1 to 0.15 percent significantly improved growth and feed conversion in juvenile shrimp.

Note: Cited references are available from the first author.

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