





# Animal byproducts can provide cholesterol for shrimp feed

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#### Not all ingredients are equal regarding sterol content



Preliminary studies indicated that various rendered products can provide sources of cholesterol to meet the dietary needs of shrimp.

One of the major concerns in shrimp nutrition is lipids nutrition, and there is ample evidence from previous studies that imbalance of sterol, particularly cholesterol, can severely reduce growth rates, molting frequency and survivals.

The ability of shrimp to synthesize sterols has been investigated by several authors, who established that shrimp are incapable of de novo sterol biosynthesis. Thus it is believed that shrimp rely upon a dietary source of cholesterol, though some studies have shown the occurrence of cholesterol synthesis in vitro by shrimp mitochondria. In addition, desmosterol, an intermediate in the de novo synthesis of cholesterol, seems to be converted into cholesterol by some species of shrimp.

While all ingredients carry some nutritional benefits, not all are created equal when it comes to sterol content. Alternative sources of sterols are emerging, and research is under way to increase the cholesterol content in poultry products for feed. Likewise, the high cost of cholesterol used in aquaculture feeds for shrimp makes it important to precisely define their requirement in order to avoid excess supplementation.

### Feed study

To help define sterol requirements for shrimp, the authors examined the sterol composition of a number of animal feed samples to determine their suitability as shrimp feed.

Lipids were extracted from each sample (Table 1) according to a modified Bligh and Dyer extraction. This involved a first step of suspending the samples in a mixture of buffer that disrupts cells. To this mixture were added water and additional chloroform. The samples then sat overnight before the lower lipid-containing chloroform layer was removed under rotary evaporation and redissolved in methylene chloride.

## Nates, Relative percentages of sterols, Table 1

	Sterols as % of Total Lipids Extracted	СН	CH1	СНО	САМ		LAN	LAN2	SIT
Blood meal	0.29	100.00							
Corn oil*		0.50			23.00	1.00			66.00
Cottonseed oil*		0.50			4.00	0.50			93.00
Feather meal	13.10	73.00	1.20	18.20	0.30		4.70	2.60	
Fish oil*		37.70			10.30	0.30			71.70
Meat and bone meal	0.42	89.30	1.00		2.60	6.00	6.10	8.50	4.50
Palm oil*		1.00			14.00	1.00			74.00
Poultry by- product meal	2.32	74.10	1.10		1.00		17.60	6.30	
Poultry by- product meal, pet grade	0.59	81.80	1.30		1.40		7.80	7.80	
Poultry by- product meal, feed grade	1.29	77.20	1.50		1.40		15.70	4.20	
Soybean oil*		0.50			20.00	3.00			53.00

Table 1. Relative percentages of sterols found in animal by-products and plant oils.

\* T. M. Jeong, 1975 CH = Cholesterol CH1 = Unsaturated Cholestrol CHO = Cholesterol CAM = Campesterol ST1 = Stigmasterol LAN = Lanosterol LAN2 = Lanosterol, Less Saturation SIT = Sitosterol

Derivatization of sterols fractions was performed according to the methodology utilized in 2002 by Jeffrey Leblond and Peter Chapman. The methylene chloride within which the lipid extract was dissolved was removed, and the lipids were saponified by heating.

After cooling to room temperature, 0.5 mL of glacial acetic acid were added, the sample was vortexed, and then 1 mL of water was added. Non-saponifiables and free fatty acids were removed with three extractions.

Sterols were characterized by examination of trimethylsilyl ether derivatives. The reagent was evaporated under a stream of nitrogen, and the derivatives were redissolved in 40 mL of 1:1 hexane/MTBE. Mass spectrometry analysis was performed using the same conditions described by Leblond and Chapman with the exception that the final hold temperature was 300 rather than 310 degrees-C.

### Results

In each animal byproduct sample, the most abundant sterol was cholesterol at levels that were never below 70 percent of the total sterols (Table 1). While only the blood meal sample possessed only cholesterol, the majority of samples possessed other sterols at low levels. From these preliminary studies, rendered products seem to be cost-effective sources of cholesterol for fish and shrimp diets.

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