

Strategies to reduce feed cost by improving gut health and nutrient utilisation of fish in aquaculture

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Feed is an important input that involves nearly 60% of the total production cost in commercial aquaculture. Hence, nutritionally balanced and cost effective diets play a large role in deciding the return from an aquaculture venture. The quality of nutrients is determined by their source, ie. the ingredients used in diet formulation which in turn determines the feed cost.

Protein is the most expensive component in compounded feed because protein-rich ingredients are typically costly and their inclusion in aquaculture diets has a significant role in overall feed costs. In aquaculture production, fishmeal is usually regarded as the main protein source in formulated feed owing to its high protein content, excellent amino acid profile, low carbohydrate level, high digestibility and its palatability. During the past decades worldwide fishmeal demand and supply has been greatly affected leading to higher costs. To find a solution, nutritionists are searching for alternative ingredients that have a reasonable price and nutritionally balanced amino acid profile to meet the requirements of cultured fish. In commercial aquaculture practices overall growth depends upon the gut health of the animal and nutrient utilisation. Hence, this article discusses the various strategies to reduce the feed cost by promoting gut health and nutrient utilisation.

Alternative sources of fishmeal in aquaculture feeds

Animal by-products, due to their high protein and low carbohydrate content, balanced amino acid profiles, lack of anti-nutritional factors and palatability can be considered as alternate feed ingredients to fishmeal. Ingredients of plant origin such as aquatic weeds, legume seeds, various oilseeds and cakes, leaf meals, leaf protein concentrates, and root tuber meals may also be used in aquaculture feeds as they also contain high protein levels and may be locally available at a cheap price.

Hurdles in utilisation of plant-based ingredients in fish feed

Some studies have shown that plant-based ingredients are able to fully or partially replace fishmeal without affecting the growth performance of farmed fish and shrimp species due to presence of high protein, with the added benefit of a low cost per kilogram of protein compared to fishmeal. However, some plant-based ingredients can be nutritionally complicated with

an unbalanced amino acid profile or low nutrient digestibility and bioavailability. They may also contain anti-nutritional factors or nonsoluble carbohydrates.

Most plant-based ingredients are deficient in some essential amino acid, such as lysine or methionine. For example corn gluten meal is an alternative protein source to fishmeal in some feeds. It has a 60% crude protein content but it is limited in lysine and contains a lot of nonsoluble carbohydrate. Soy protein is also an important plant-based protein source, but is deficient in some essential amino acids, essential minerals and contains anti-nutritional factors such as phytic acid, which is tightly bound to essential minerals such as P, Ca, K, Cu, Mg, Fe, Zn and protein. Phytic acid mainly reduces the bioavailability of phosphorus. Additionally, other anti-nutritional factors such as tannins and protease inhibitors hinder protein digestibility because they reduce the proteolytic enzyme activity (these inhibitors mainly act by binding with chymotrypsin and trypsin). Tannins are present in a wide variety of plant ingredients.

Lectins or haemagglutinins cause a reduction in absorption of nutrients from the gut and alimentary canal of organisms. Gossypol is also an important anti-nutritional factor present in cotton seed, which forms a complex with protein and may lead to deficiency of methionine and lysine amino acids.

Oxalates forms complexes with some essential minerals like calcium. Oil cakes including rapeseed and mustard contain glucosinolates, which inhibit the uptake of iodine by the thyroid gland.

Mimosine is another anti-nutritional factor present in ipil-ipil. Mimosine is an unusual amino acid, structurally it is like tyrosine and is an antagonist to other amino acids and affects the thyroid gland.

Non-starch polysaccharides

Non-starch polysaccharides (NSP) are polymers of hexoses and pentoses (galactose, glucose, xylose, mannose, etc) and are not easily digestible in fish. NSP comprises up to 90% of the plant cell wall (cellulose, hemicelluloses and pectins). Cellulose is a basic structural component of the cell wall. Xylan is the most common hemicellulose and represents the major non-cellulosic cell wall polysaccharide in plants. Cellulase enzyme breaks β -(1,4) linkages in cellulose, but since fish lack this enzyme the utilisation of cellulose is low in most fish.

Some conventional methods such as heat treatment, soaking in water, germination and de-hulling methods can effectively inactivate anti-nutritional factors in plant feed stuffs but

increase cost and time required for feed preparation. Some anti-nutritional factors are heat stable. So the following sustainable approaches are proposed to effectively reduce the problems of plant-based ingredients and finally reduce feed costs in aquaculture.

Sustainable approaches to overcome the hurdles

Approaches that can be used to improve nutrient utilisation and reduce feed cost include:

- Probiotic approach - to modulate the gut microflora using selected bacteria.
- Prebiotic approach - specific nutrients promoting the development of selected bacterial strains.
- Supplementation of exogenous enzyme - to improve digestibility.
- Supplementation of essential nutrients - to overcome specific nutrient deficiencies of ingredients (such as essential amino acids, vitamins, minerals and fatty acids).
- Supplementation of chemo-attractants - stimulate appetite and palatability.
- Other feed additives (acidifiers and mycotoxin binders) – digestibility enhancers.

Fish gut microbiota.

The fish gut contains a microbial community of aerobic and anaerobic bacteria, yeast and fungi. Fish gut bacteria can be divided into autochthonous (able to colonise in the gut) and allochthonous (considered to be free living). The microbial composition largely depends on the nutritional status of the fish. Mostly, protease enzyme-producing bacteria are present in carnivorous fish, whereas amylase- and cellulase-producing bacteria are present in herbivorous fish. Autochthonous bacteria produce endogenous digestive enzymes that are helpful for the digestibility of feedstuff.

Many studies have isolated useful enzyme producing bacteria from the different fish species. Cellulose-degrading microorganisms convert cellulose into a simple molecule of glucose. Phytase-degrading bacteria increase the bioavailability of

phosphorus and other minerals. Tannase enzyme degrades tannins into inactive forms of tannic acid and xylanase enzyme hydrolyse xylan into xylase molecules.

Gut microbiota as a bio control agent: A probiotic approach

Probiotics are defined as live microorganisms, when administered in sufficient amounts, that confer a health benefit to the animal. So the use of enzyme-producing indigenous microbiota can be a form of probiotics. Probiotic organisms may confer health benefits to the host by improving feed digestibility through the supply of exogenous enzymes and degradation of complex components of the feed that are otherwise hard to digest. Probiotics may produce antagonistic compounds against pathogens, so they may be used as alternative to antibiotics. Moreover, probiotics are able to be grown in low pH and bile salts affect the fish gut.

Fermentation: An approach for bio-processing of feedstuffs

Fermentation is based on use of microorganisms that can exhibit a beneficial role, if reared under specific conditions. Fermentation can increase the crude protein and decrease the content of crude fibre, anti-nutritional factors and toxic contents in feedstuffs. Fermentation improves the availability of vitamins, protein solubility and amino acid profile as well as increasing the palatability. Bacterial strains (*Bacillus* sp., *Enterococcus* sp., *Lactobacillus* sp.), yeast (*Saccharomyces* sp.), and fungal cells (*Aspergillus* sp) can be used in fermentation of feedstuffs. Bacterial cells obtain energy by converting carbohydrates (glucose or lactose) into lactic acid. Yeasts degrade complex carbohydrate into glucose, ethanol and carbon dioxide.

Supplementation of exogenous enzymes

Supplementation of exogenous enzymes enhances the digestibility of plant feedstuffs through hydrolysis of complex carbohydrates and inactivation of anti-nutritional factors. So they act to increase the nutrient availability and digestibility of proteins, carbohydrates and phosphorus content. Carbohydrase includes all enzymes that catalyse a reduction in the molecular weight of polymeric carbohydrates, which aids easy digestion. In fish feed, more than 80% of supplemented carbohydrase are xylanase and glucanase. Other commercially available carbohydrase include α amylase, β mannanase, α galactosidase and pectinase. When the presence of specific digestive enzymes seems to be very low or non-existent in fish, supplementation with exogenous enzymes will increase the bioavailability of

Table 1. Non starch polysaccharides present in plant feed stuffs (Source: Sinha et al., 2011).

Category	Monomeric residue	Linkage	Plant feedstuffs
Cellulose	Glucose	β -(1 \rightarrow 4)	Cereals (barley, corn, wheat) and legumes (soybean, cottonseed, rapeseed/canola, lupin)
Arabinoxylans	Arabinose & xylose	β -(1 \rightarrow 4) linked xylose units	Cereals (rye, rice, barley, corn, wheat, oat, sorghum)
Mixed linked β -glucans	Glucose	β -(1 \rightarrow 4) and β -(1 \rightarrow 4)	Barley & oats
Arabinans	Arabinose	α -(1 \rightarrow 5)	Cereal co products
Galactans	Galactose	β -(1 \rightarrow 4)	Sugar bean meal, Sugar beat pulp
Arabinogalactans (Type-I)	Arabinose & galactose	β -(1 \rightarrow 4)	Grain legumes
Arabinogalactans (Type-II)	Arabinose & galactose	β -(1 \rightarrow 4)	Rapeseed cotyledon

carbohydrate and minerals. Mostly, enzymes are inactivated at high temperature (95°C). So, exogenous enzymes are mixed with oil and sprayed onto the feed.

Acidifiers

Generally, agastric fishes do not have an acidic pH due to the absence of a stomach. Most enzymes like phytase act at an acidic pH. To overcome such problems acidifiers are used to enhance the activity of enzymes, which in turn improves digestibility. Many organic acid feed additives are supplemented to the diets because of their antimicrobial effects. Mostly, the acidifiers used are organic acids (short chain fatty acids). Organic acids are low molecular weight aldehyde-containing compounds with one or more – COOH groups. Organic acids, such as acetic, butyric, citric, formic, lactic, propionic, malic, and sorbic acids and their salts have been used as acidifiers in animal feeds (NRC, 2011). Organic acids are available on the market in a variety of forms such as adsorbates (liquid acids or mixtures of acids adsorbed onto a solid), salts (usually solids, except for ammonium propionate and ammonium formate which are liquid). Organic acids are applied directly into feedstuffs and compound feed. Liquid acid blends are sprayed onto the feed, whereas solid acids and acid salts are added directly or via special pre mixtures. The mode of action of organic acids in the intestinal tract involves two different actions:

- The pH-decreasing action of organic acids in the stomach and small intestine through delivery of H⁺ ions contributes to an improved activity of digestive enzymes.
- They inhibit the growth of gram negative bacteria through the dissociation of the acids and production of anions in the bacterial cells.

The use of acidifiers can be an efficient tool to achieve sustainable, economical and safe fish and shrimp production.

Phytogenics

Phytogenics are products originating from the leaves, roots, tubers, fruits or spices of herbs and other plants. Phytogenics are available either in solid, dried and ground forms, and as extracts or essential oils. The active ingredients of these products (eg. phenolics and flavonoids) can lead to stimulation of the appetite, increased antimicrobial action, direct reduction of gut bacteria, stimulation of gastric juices, enhancement of immune system, anti-inflammatory responses or may have antioxidant properties. The antimicrobial mode of action is considered to arise mainly from the potential of hydrophobic essential oils to intrude into the bacterial cell membrane, disintegrate membrane

structures, and cause ion leakage. The garlic extract allicin (diallyl thiosulfinate) is used for bacterial disease control and immunostimulation of cultured fish. Essential oils extracted from rosemary also seem particularly interesting due to their high concentration of components such as carnosol and carnosic acid, which have high antioxidant properties. In addition, peppermint and cinnamon also seem to be possible candidates to be used as phytochemical feed additives in aquatic species to improve growth performance, fish health status and to reduce microbial challenge in the gut.

Mycotoxin binders

In commercial aquaculture feed production the usage of plant-based proteins instead of fishmeal, leads to contamination by mycotoxins. Mycotoxins are secondary metabolites produced by different species of fungi that have the potential to reduce the growth and health status of aquatic organisms. There are a number of options to prevent or reduce the risk of mycotoxin contamination such as careful selection of raw materials, maintaining good storage conditions for feeds and raw materials, and using feed additives. Aluminum silicates, clay and zeolitic materials, are the most commonly applied group of mycotoxin binders.

Chemoattractants

Plant feeds generally have lower palatability than fishmeal. So, chemoattractants can be used to increase feed consumption by fish. Commercially, both natural and synthetic chemoattractants are used. Krill meal, anchovy fishmeal, byproducts of squid meal, fish solubles, fish hydrolysate (tuna hydrolysate, squid hydrolysate), poultry meal, blood meal, hydrolysed feather meal and some herbal plants are used as natural chemoattractants in fish feed formulations. A mix of free amino acids (alanine, arginine, glutamine, glycine, isoleucine, serine, taurine, betaine, proline), nucleotides and nucleosides are examples of synthetic chemoattractants also used in feeds.

Conclusion

Formulated feed is very important in aquaculture production. In formulated feeds fishmeal is usually considered the primary source of protein and it is the main factor in overall feed production cost. Plant-based feedstuffs as an alternative to fishmeal have been a focal area of research globally but the presence of deleterious anti-nutritional factors associated with plant-based ingredients and nutritional deficiencies have limited their use. The various strategies discussed in this article can be adopted to bring down feed costs and improve nutrient utilisation in fish.

Table 2. Type of commercial feed enzymes and target substrates.

Enzymes	Target substrate	Target feed stuffs
β-glucanases	β-glucan	Barley, oats and rye
Xylanases	Arabinoxylans	Wheat, rye, triticale, barley, fibrous plant materials
α-galactosidases	Oligosaccharides	Soybean meal, grain legumes
Phytases	Phytic acid	All plant-derived ingredients
Proteases	Proteins	All plant protein sources
Amylase	Starch	Cereal grains, grain legumes
Lipases	Lipids	Lipids in feed ingredients
Mannanases, cellulases, hemicellulases, pectinases	Cell wall matrix (fibre components)	Plant-derived ingredients, fibrous plant materials