PLANT PROTEIN INGREDIENTS FOR AQUAFEEDS

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Introduction

Aquaculture feed ingredients tend to be mostly by-products of processing or milling industries, but also consist of natural products. In commercial formulation of diets, these ingredients are included and substitutions made within mixtures in accordance with market price, local availability, digestibility, and nutrient composition. Basically, the concept is to use available ingredients in the most economical way to provide the essential nutrients required by the aquatic animal for cost-effective production. Different proportions of less expensive ingredients can often be combined to achieve the nutrient balance of more expensive ones. However, it is also necessary to consider factors such as the quality, palatability, digestibility, and functional properties of ingredients as well as the possible content of anti-nutritional components that are known to negatively affect the growth and health of fish.

The purpose of this paper is to briefly review published information about five of the most commonly available feed ingredients of plant origin, and to provide guidelines for standards and usage of these ingredients in aquaculture feeds.

Ingredients of Plant Origin

Plant protein supplements, cereal grains, and grain by-products are widely used in feeds for aquaculture species. Global availability and relatively low cost are attributes of these feedstuffs. Properly processed plant products and by-products generally also have high protein digestibility. They can often be used in combination to replace more expensive ingredients such as fishmeal (Table 1). Without exception, however, every ingredient of plant origin has some component or other factor that requires consideration or limits its use in aquaculture feeds.

Table 1. Combination of protein sources to balance amino acids

Ingredients	Protein (%)	Methionine (%)	Cystine (%)	Lysine (%)	Met & Cys: Lys Ratio
Soybean Meal	47	0.7	0.7	3.2	0.4
Corn Gluten Meal	60	1.9	1.1	1.0	3.0
Soybean Meal (90%) & Corn Gluten (10%)	49	0.8	0.8	3.0	0.5
Herring Meal	70	2.2	0.7	5.7	0.5

Soybean Meal

Among ingredients of plant origin, the relatively high crude protein contents and well-balanced amino acid profile of soy protein, as well as reasonable cost, have made soybean meals important ingredients in aquaculture feeds. The steady supply, wide availability, and consistent composition of various soy products with respect to both nutrient composition and physical characteristics in feed processing are other positive factors that have contributed to their widespread use.

Meal Products

On a global basis, heat processed full-fat soybeans, mechanically extracted soybean cake, solvent extracted soybean meal, dehulled solvent extracted soybean meal, and soy protein concentrate are the most commonly used soybean products in feeds for aquaculture species. These products are produced using different methods of processing whole beans to extract oil, control protein content, and/or reduce the activity of anti-nutrients. The proximate composition of these soybean products is presented in Table 2.

Table 2. Nutrient composition of soybean products commonly used in aquafeeds

Description – Soybean Internat. Feed. No.	Seeds, heat processed, full-fat (1) 5 – 04 – 597	Soybean meal, mech. extd. (2) 5 – 04 - 600	Soybean meal, solv. extd. (1) 5 – 04 - 604	Soybean meal, w/o hulls, solv. extd (1) 5 – 04 - 612	Soy Protein Conc. (1) -
Dry Matter (%)	90	90	89	90	92
Crude Protein (%)	35.2	42.9	44.0	48.5	63.6
Ether Extract (%)	18.0	4.8	1.5	0.9	0.5
Crude Fiber (%)	5.0	5.9	7.3	3.4	4.5
Ash (%)	4.5	6.0	6.3	5.8	-

(1) Adapted from National Research Council, 2011

(2) Adapted from National Research Council, 1982

Processing of full-fat soybeans is done either by extrusion through a high-temperature-short time expander, or roasting whole in a fluidized bed of hot air (Figure 1). When ground, beans processed by the roasting method form a meal that has functional properties similar to solvent extracted soybean meal. With this type of meal it is possible to formulate pelleted diets containing high levels of fat. Meals from both heat treatment methods can be effectively used in formulated diets for a wide variety of fish species (Lim and Akiyama, 1989). Full-fat soybeans, when properly heat-treated, have been shown to be an excellent source of protein and energy in diets for trout (Smith, 1977), catfish (Saad, 1979) and tilapia (Tacon et al., 1983).

Mechanically processed meals can also be produced in two ways. By the old method, soybeans are crushed into flakes, which are subjected to steam cooking. The hot, wet soy flakes are then spread in layers between heavy cloth and placed in a press, where as much of the oil as possible is squeezed out by pressure. The resulting cakes are broken into smaller pieces and sold in that form, or ground into a granular meal. The newer, expeller method does the same job of extracting oil from the beans with moist heat and pressure, however, it is done in a continuous process with a screw press. With both mechanical oil extraction methods, the meal retains approximately 5% fat.

Solvent extraction is the most widely employed method of producing soy oil and meals (Figure 2). This process utilizes a fat solvent, usually hexane, in which dehulled, steam conditioned soy flakes are soaked and counter-currently washed with clean solvent to reduce the oil content to less than 1%. After the oil is extracted, the residual meal is heated with steam to volatilize the remaining solvent and may be further toasted to denature growth-inhibiting proteins. The meal is then dried, cooled, and ground to a uniform particle size. Toasted and ground hulls, removed at the beginning of the extraction process may be added back to the meal to produce a higher fiber, lower protein product.

Figure 1. Roaster for full-fat soybeans

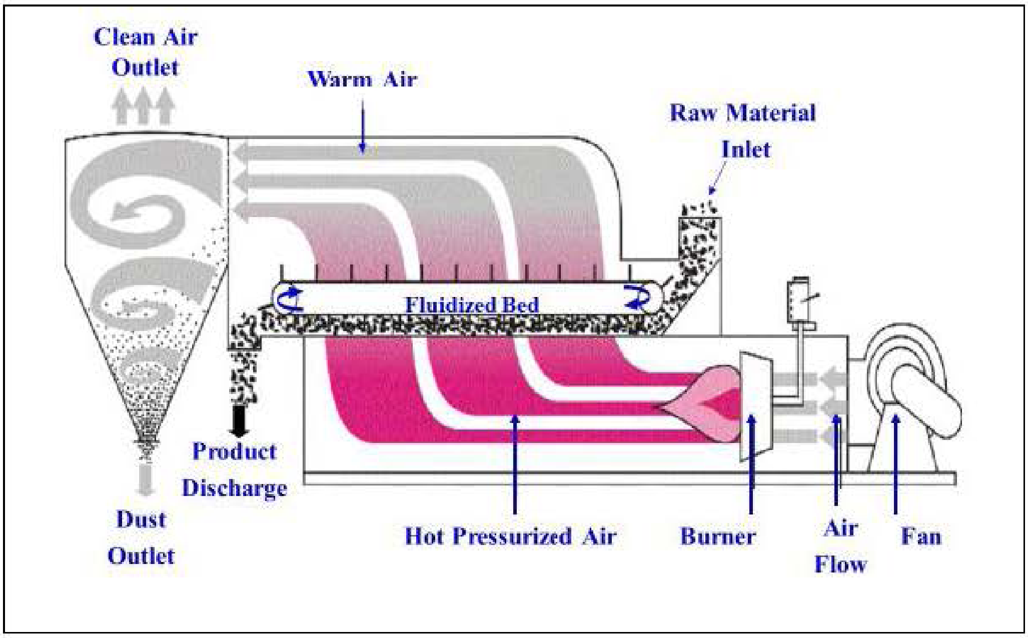
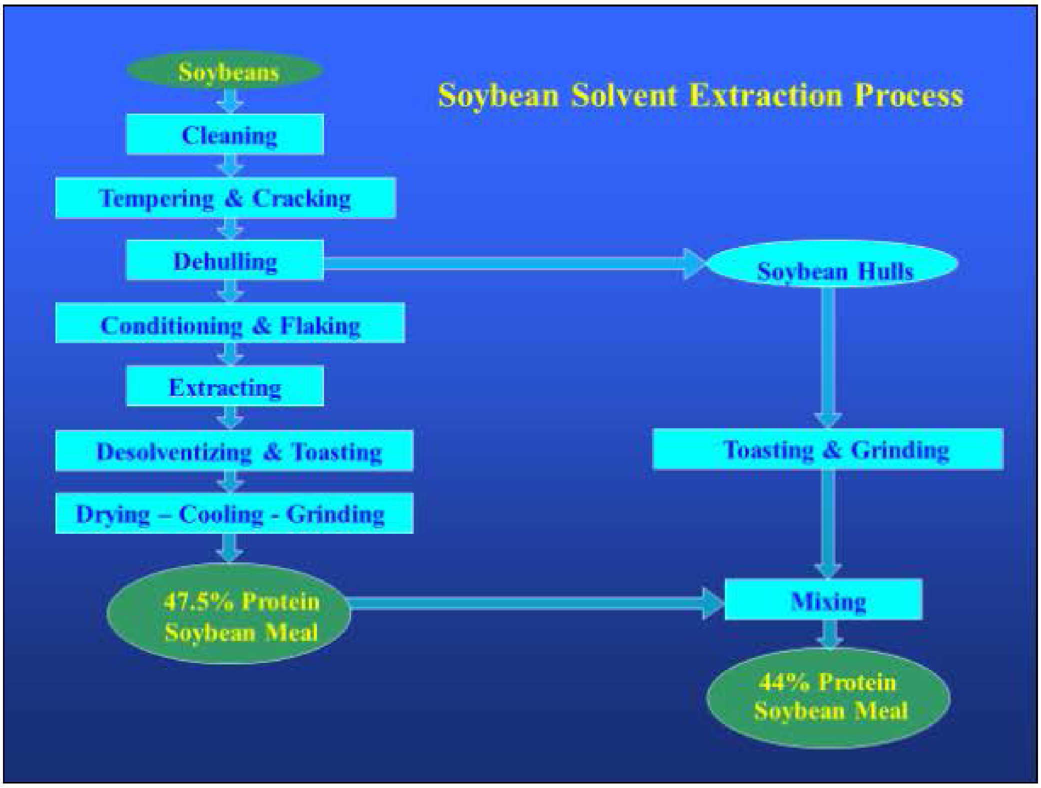
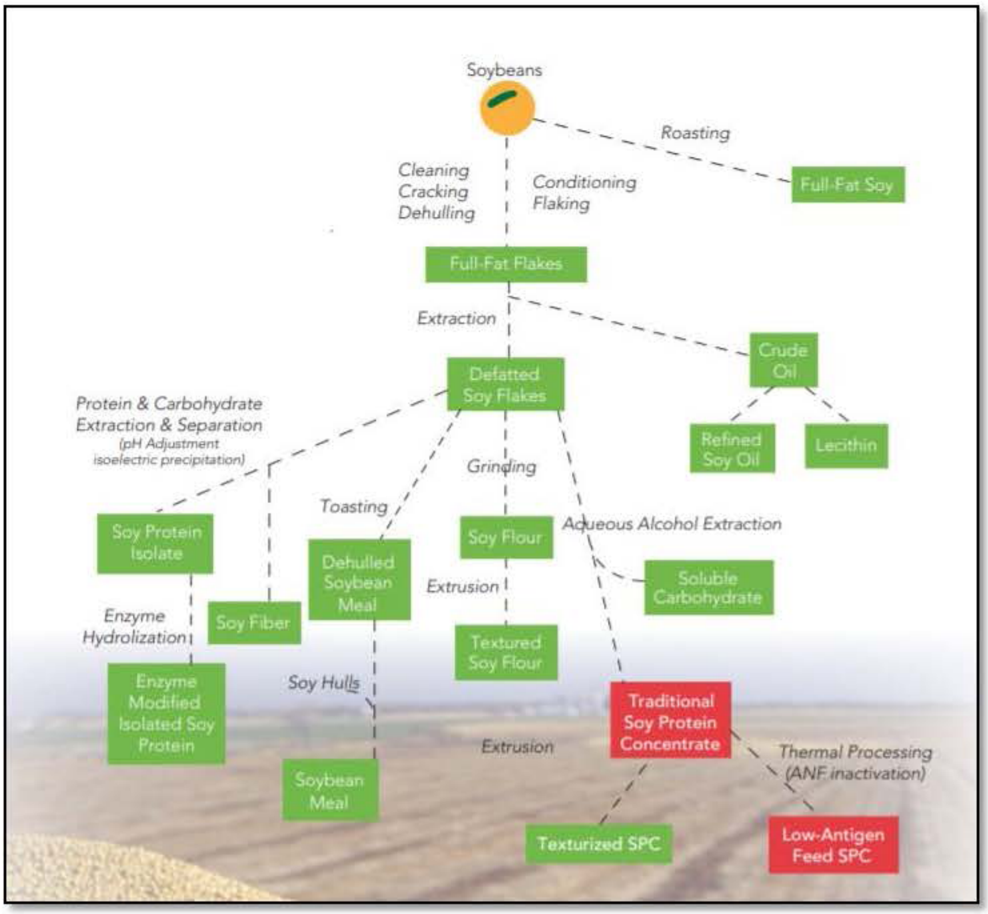


Figure 2. Soybean solvent extraction process



The process of manufacturing soy protein concentrate (SPC) begins with the same preparatory steps as in making solvent extracted soybean meal (Figure 3), however in the case of making SPC the defatted white flakes are treated with aqueous alcohol to remove soluble carbohydrates and to significantly reduce the anti-nutritional factors: lectins, trypsin inhibitors, glycinin, B-conglycinin, saponins, and oligosaccharides. In that process, the protein content of SPC is concentrated up to a range of 63-65%. The resulting traditional SPC is heat-treated to further reduce the anti-nutritional factors to produce low-antigen, feed grade SPC which is suitable for use in a wide variety of aquafeeds.

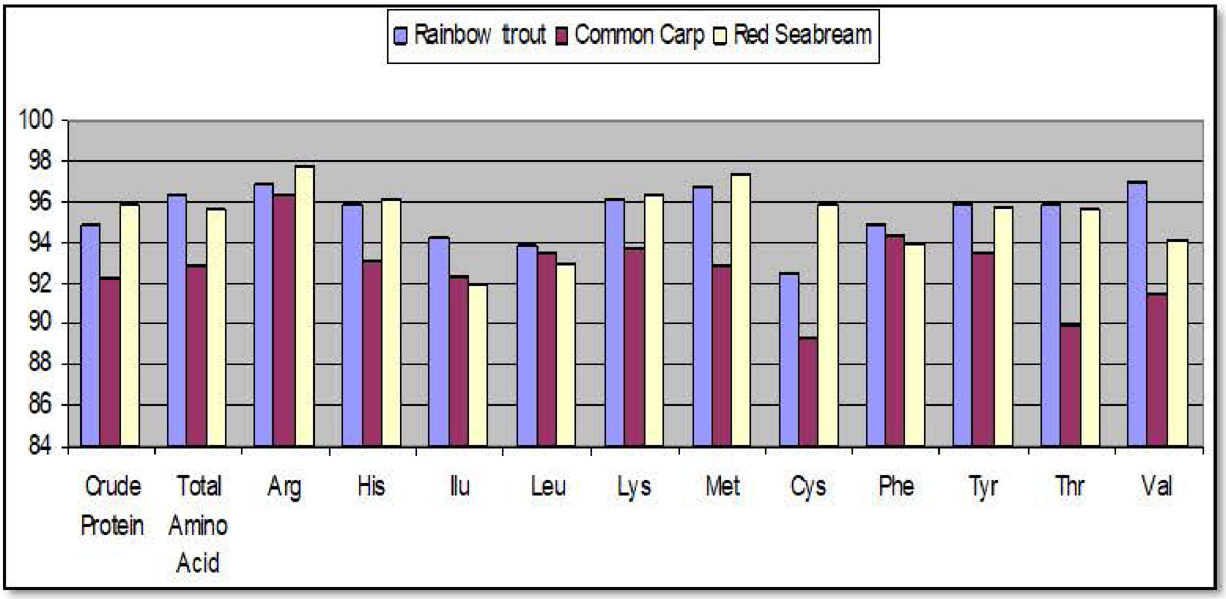
Figure 3. Process for manufacturing soy protein concentrate (USSEC, 2008)



Species Differences in Digestibility

In formulating diets containing soy protein it is important to note that research has shown that the digestibility of protein and amino acids from soybean meal is different in different species of fish (Figure 4). Yamamoto and coworkers (1998) found the digestibility of crude protein and total amino acids was roughly similar in two carnivorous species, rainbow trout (*Onchorynchus mykiss*) and red seabream (*Chysophrys major*), even though the water temperatures for optimum growth of these species are very different. However, these were higher than digestibilities measured in the common carp (*Cyprinus carpio*), which is an herbivorous fish without a true acid stomach. They also found that the digestibility rates for the individual amino acids were completely different among the species tested. Separate research with the omnivorous channel catfish (*Ictalurus punctatus*) has shown the digestibility of protein from soy to be among the highest for all feed ingredients typically used for this species (Wilson and Poe, 1985). NRC (2011) listed apparent digestibility of protein of soybean meal for 17 species of freshwater and marine fish in the range of 86 – 95%, with several outlying values, and for Penaeid shrimp in the range of 89-97%. In the same publication, apparent digestibility of protein of soy protein concentrate was listed in the range of 87-100% for five species of fish, and 93% for Penaeid shrimp. These reported research findings emphasize the need for more nutrient digestibility data for each fish species to avoid errors made by applying digestibility data across species.

Figure 4. Percent digestibility of crude protein and essential amino acids from solvent extracted soybean meal in fingerling rainbow trout, common carp, and red seabream (Yamamoto et al., 1998)



Anti-Nutritional Factors

Among the critical considerations that must be made when using soybean meals in feed is the fact that raw soybeans contain several anti-nutritional factors know to affect negatively the growth and health of fish. Some of these can be inactivated or eliminated by heat treatment of the meal. These include protease inhibitors, hemagglutinins, goitrogens, and phytates (Table 5, from Liener, 1980).

The only heat-labile anti-nutritional factor of any practical significance in fish nutrition is trypsin inhibitor. If sufficient quantities of this enzyme are present in the soybean portion of the diet, it can tie up the trypsin required for complete digestion of all dietary protein. Heat treatment of the meal denatures trypsin inhibitor enzyme, effectively inactivating it. The amount of active trypsin inhibitor is related to the type of heat treatment as well as the temperature and duration of exposure.

The optimum conditions for heat treatments, as well as the best chemical means of determining the adequacy of heat treatment, are constantly being revised. However, the most frequently used chemical criteria are urease activity, trypsin inhibitor value, and protein solubility index. Values for these test criteria for soybean products suitable for aquaculture species are, 1-3 mg trypsin inhibitor activity per g of sample, urease increase in pH between 0.0 and 0.23, and protein solubility index of 75-85%.

Table 3. Anti-nutritional factors in soybeans

Heat-Labile	Heat-Stable
Protease Inhibitors	Oligosaccharides
Hemagglutinins	Non-Starch Polysaccharides
Goitrogens	Estrogens
Phytates	Allergens

Lim and Akiyama (1989) caution that the most accurate means for assessing the nutritional value of soy meals are biological indicators such as digestibility values, growth, feed utilization efficiency and sub-clinical (presumably histological) abnormal signs. This is because some of the anti-nutritional components of soybeans are not eliminated by heat. These include oligosaccharides, non-starch polysaccharides, estrogens and antigenic proteins (Liener, 1980). Different species of fish apparently have different levels of tolerance or sensitivity to these heat-stable components (Storebakken et al., 1999).

The carbohydrate portion of soybeans includes the oligosaccharides sucrose, raffinose, and stachyose. While sucrose is digestible by fish, the other two oligosaccharides are not. Their presence in the intestinal contents increases the osmotic pressure of the fluid and thereby restricts the absorption of water. These indigestible oligosaccharides do not pose any problems in freshwater fish, which are constantly excreting water to maintain the osmotic pressure of their body fluids in a hypo-osmotic environment. In marine species, however, it is believed that the reduced absorption of moisture from the intestinal contents is a source of osmoregulatory stress when the fish are raised in seawater.

The nutritional actions of non-starch polysaccharides are not fully understood. These compounds are known to cause increased viscosity of the intestinal contents in poultry. One published research report on non-starch polysaccharides in diets for Atlantic salmon (Refstie, et al., 1999) attributed a trend of reduced digestibility of fat and protein to the possible effect of increased viscosity of intestinal contents on diffusion and mixing of digestive enzymes. However, this observation has never been reported in studies with freshwater fish. It may be that non-starch polysaccharides simply have the same effect as oligosaccharides on the water balance in fish raised in a marine environment.

Estrogenic and allergic effects of soy components in fish appear to be highly species specific. Soy isoflavones have been shown to cause increased plasma concentrations of sex hormones in immature sturgeon. However, this effect has never been reported in any species of bony fish.

Soy components in full-fat and fat-extracted soybean meals cause morphological changes in the mucosa of the distal intestine of salmon and other marine species. Bureau et al., (1998) and Knudsen et al., (2007) showed that soy saponins increase the intestinal epithelial permeability, and can interact with unidentified components in legumes to induce an inflammatory reaction. As the soy saponins are eliminated in the process of manufacturing soy protein concentrate, SPC can be included in diets of salmon and other marine species at considerably higher levels than soybean meals without causing inflammatory reactions in the intestines of these species.

Formulation Recommendations

Research on the use of soybean protein in fish feeds has been conducted for more than 40 years and with many aquaculture species. However, unlikethe type of research that has been done with poultry and swine, the extreme number of variables involved has complicated this body of work on fish. Feed formulation and ingredient differences, changes in feed manufacturing technology, different environmental conditions, and extreme differences in genetic stocks within each species all combine to make it impossible to prescribe absolute usage guidelines for soybean meals in aquaculture feeds. The following table presents conservative recommendations for the maximum amounts of soy nitrogen free extract (soy NFE) that could be used in feeds for several of the most common species in aquaculture. Soy NFE is used as a rough approximation of the amount of heat-stable anti-nutritional factors in different soy products, and is easily calculated for each soy product in stock in feed mills by the equation: % NFE = % dry matter – (% crude lipid + % crude protein + % ash + % crude fiber).

Table 4. Maximum inclusion rates of Soy NFE in feeds for aquaculture species

Species	Max. Soy Nfe (%)
Carp	16 - 17
Tilapia	16 - 17
Channel Catfish	16 - 17
Rainbow Trout	7.5 - 8.5
Chinook Salmon	0
Coho Salmon	6.0 - 6.5
Atlantic Salmon	3.0 - 3.5
Red Drum	6.0 - 6.5
Striped Bass	7.5 - 8.5
Red Seabream	7.5 - 8.5
Japanese Eel	6.0 - 6.5
Marine Shrimp	13.5 - 14.5

Cottonseed Meal

Cottonseed is perhaps the second most abundant source of plant protein in the world. As with soybean, this oil seed is processed in several different ways to yield cottonseed oil and a variety of meal products. All of the meals are high in protein and appear to be palatable to most species of fish. In high cotton production areas, cottonseed meals are generally less expensive per unit of protein than soybean meals. However, the use of cottonseed meal products in feeds for aquaculture species has been limited. The primary reason for this is that cottonseeds contain anti-nutritional components, free gossypol and cyclopropenoid fatty acids, which are harmful to fish when present in sufficient quantities. Cottonseed meals are also low in lysine content and high in fiber. In spite of these inherent negative characteristics, good quality cottonseed meals can be effectively formulated into aquaculture feeds when economic conditions favor their use.

Meal Products

The basic processes of oil extraction from cottonseed are mechanical extraction by screw press, mechanical extraction followed by solvent extraction, and direct solvent extraction. The resulting meals have different nutrient compositions. Table 5 illustrates the proximate compositions of four of the most commonly produced cottonseed meals.

Table 5. Nutrient composition of cottonseed meals commonly used in fish feeds

Description – Cotton	Seeds, meal mech. Extd. (1)	Seeds, meal solv. extd. (2)	Seeds, meal prepressed solv. extd. (1)	Seeds w/o hulls, meal prepressed solv. extd. (1)
Internat. Feed No.	**5 – 01 – 617**	**5 – 07 – 872**	**5 – 07 - 873**	**5 – 07 - 874**
Dry Matter (%)	93.0	92.0	91.0	90.0
Crude Protein (%)	41.0	41.7	44.7	48.6
Ether Extract (%)	4.6	1.8	1.6	1.2
Crude Fiber (%)	11.9	11.3	11.1	7.9
Ash (%)	6.1	6.4	6.1	6.4

(1) Adapted from National Research Council, 1982
(2) Adapted from National Research Council, 2011

Nutrient Composition

The high protein and relatively low fiber content of dehulled, prepressed, solvent extracted meal make it the preferred cottonseed meal product for use in fish feeds. However, prepressed solvent extracted meal made from whole seeds can also provide economic advantages in some formulations. The primary consideration for use should be the contribution to providing the required levels of essential amino acids in the diet.

Cottonseed protein compared to that of soybean is very high in arginine (Table 6). However, it is severely deficient in lysine and slightly deficient in isoleucine and the sulfur containing amino acids, methionine and cystine. The true availability of each of the essential amino acids, as determined in channel catfish (Wilson et al, 1981), have also been found to be lower in cottonseed meal than in soybean meal. More recent data (IAFFD, 2020) confirms that availability of essential amino acids by fish, in general, is lower in cottonseed meal than in soybean meal. The IAFFD database shows that the digestibility of essential amino acids contained in cottonseed is on average about 11% lower than that of soybean meal.

Anti-Nutritional Factors

Utilization of cottonseed meal in feeds for aquaculture species is limited by the presence of gossypol. This is a yellow pigment, which is a highly reactive polyphenolic dinaphthaldehyde compound found in the oil gland of cottonseed. Gossypol, in its free (unbound) form, causes anorexia, slow growth, and increased fat deposition in liver tissue when fed to fish in excess (Wood and Yasutake, 1956). Free gossypol has also been reported to increase the incidence of and growth of aflatoxin-induced liver tumors in rainbow trout (Sinnhuber et al., 1968). Clinical symptoms of gossypol toxicity apparently occur in all fish, although research reports indicate considerable species variation in sensitivity.

Table 6. Comparison of the composition and true availability of essential amino acids in cottonseed and soybean meals

Essential Amino Acids	Cottonseed Meal (1) Composition (3) (%)	Cottonseed Meal (1) Availability (4) (%)	Soybean Meal (2) Composition (3) %	Soybean Meal (2) Availability (4) %
Arginine	10.0	90.6	7.4	96.8
Histidine	2.6	81.6	2.7	87.9
Isoleucine	3.5	71.7	5.4	79.7
Leucine	5.6	76.4	7.8	83.5
Lysine	3.8	71.2	4.6	94.1
Methionine	1.4	75.8	1.4	84.6
Cystine	1.7	-	1.5	-
Phenylalanine	5.2	83.5	5.6	84.2
Tyrosine	2.3	73.4	2.6	83.3
Threonine	3.2	76.7	4.1	82.2
Tryptophan	1.3	-	1.4	-
Valine	4.6	76.1	5.6	78.5

(1) Cotton, seeds, meal solvent extracted. IFN 5 - 07 - 872.
(2) Soybean, seeds without hulls, meal solvent extracted. IFN 5 - 04 - 612.
(3) Expressed as percentage of protein, data adapted from National Research Council, 2011.
(4) Determined using channel catfish (Wilson et al, 1981).

Rainbow trout (*Oncorhynchus mykiss*) fed diets containing 0.025% gossypol acetate for 18 months were found to be capable of maintaining normal growth and feed conversion, although free and bound gossypol accumulated in the fish liver tissue (Roehm et al, 1967). Other research with rainbow trout showed 0.03% dietary free gossypol suppressed growth (Herman, 1970). In the same study, levels greater than 0.05% lowered the hematocrit and hemoglobin levels in the blood, and caused necrotic changes and ceroid pigment deposition in the liver.

Channel catfish (*Ictalurus punctatus*) were found to grow normally when fed a diet containing 0.09% free gossypol from cottonseed meal (Dorsa et al., 1982). When the dietary level of free gossypol reached 0.12%, growth rate was reduced. Gossypol concentrations increased in liver, kidney, and muscle tissue as dietary free gossypol increased.

Tilapia (*Oreochromis aureus*) were reported to tolerate dietary levels of gossypol up to 0.18% (Robinson et al, 1984). However, growth rates of fish fed the test diets containing graded levels of gossypol from cottonseed meal were not as good as those of fish fed soybean meal-based diets.

The chemical characteristic of gossypol that is most responsible for limiting cottonseed meal use is that it readily binds to protein. When pigment glands in the cottonseed are disrupted during processing, free gossypol binds to the epsilon amino group of lysine in the seed protein. Proteolytic enzymes can not release gossypol-bound lysine. The percent of available lysine, which is already the most limiting amino acid in cottonseed meal protein, may be reduced below acceptable levels.

Another characteristic of cottonseed is its high susceptibility to molding and the subsequent formation of aflatoxins. Rainbow trout are particularly sensitive to these carcinogenic metabolites (Ashley, 1972 and Friedman and Shibko, 1972). Consumption of only 0.5 mg of Aflatoxin B1 per kg of body weight causes mortality within 3 to 10 days. Feeding aflatoxin-contaminated feeds with as little as 0.1 to 0.5 ppb results in hepatomas after 4 to 6 months. Other aquatic species, such as coho salmon (Ashley, 1972), catfish (Jantarotai and Lovell, 1991) and shrimp (Lightner, 1998 and Ostrowski-Meissner et al, 1994), are believed to be more tolerant, though similarly affected.

Formulation Recommendations

Cottonseed meals that have been processed by prepressing and solvent extraction make the best choice for use in feeds for aquaculture species. Research on the use of both whole and dehulled, prepressed, solvent extracted meals has been conducted mostly with salmon, trout, and catfish. Based on reports on the complete volume of work with these species, it appears that the relatively high fiber and low available lysine levels in cottonseed meal products limits economical use in commercial fish fees to no more than 15-20 percent. It is best not to use cottonseed in diets for broodstock of any species, because of the potential for prolonged feeding to cause accumulation of high tissue levels of gossypol. Hertrampf and Piedad-Pascual (2000) recommend that glandless cottonseed meal should be used whenever available, and that it should not be used at levels above 10% for marine shrimp. Finally, precautions should always be used to avoid the use of any cottonseed meal containing aflatoxins.

Rapeseed and Canola Meals

Oil seeds of the genus *Brassica*, collectively known as rapeseed, are cultivated as a source of oil and protein in many areas of the world where the climate is cool and the growing season is short. Rapeseed meals, resulting from various oil extraction processes, have relatively high fiber levels, but protein contents range from 35-40 percent. More importantly, the amino acid profile of the protein is similar to that of soybean. These nutrient characteristics of rapeseed meals make them attractive as a protein supplement in animal feed. However, use of rapeseed meals for monogastric animals has been severely limited by the existence of two problematic components. First, and most importantly, meals from traditional rapeseed contain 3-8% glucosinolate compounds, which interfere with thyroid function. Secondly, residual oil in the meal contains 25-55% erucic acid, which is known to cause cardiac lesions in rats and pigs.

During the 1970's, plant geneticists in Canada developed two new varieties of rapeseed from *Brassica napus* and *B. campestris* species. The new "canola" varieties are lower in both glucosinolates and erucic acid. By definition, canola meals contain less than 2% erucic acid in the oil fraction and less than 30 µmoles of glucosinolates per gram of air-dried, oil-free meal (AAFCO, 1998). Most research on use of rapeseed in feeds for aquatic species has been subsequently conducted only with canola meals. All of the information that follows was summarized from published data from this research with canola meals.

Meal Products

The basic canola meal products are derived by either direct solvent or prepress solvent extraction processes. Both processes are similar to those used to make soybean and cottonseed meals. The proximate compositions of these canola meal products are presented in Table 7.

Table 7. Nutrient composition of canola meals used in fish feeds

Description – Canola Internat. Feed No.	Seeds, meal solvent extracted (1) 5 - 03 - 871	Seeds, meal, prepressed, solvent extracted (2) 5 - 08 - 135
Dry Matter (%)	93.0	92.0
Crude Protein (%)	38.0	40.5
Ether Extract (%)	3.8	1.1
Crude Fiber (%)	11.1	9.3
Ash (%)	6.8	7.2
Nitrogen Free Extract	33.3	33.9

(1) Adapted from National Research Council, 2011
(2) Adapted from National Research Council, 1982

Nutrient Composition

Canola meals contain only moderate levels of protein (Table 7). The amino acid pattern is reasonably attractive for use in fish feeds (Table 8). Compared to soy protein, however, it is low in almost all of the amino acids. The percentages of true availability of essential amino acids, as determined in rainbow trout (Higgs et al, 1994), are also quite a bit lower compared to those in soy protein. More recent data (IAFFD, 2020) confirms that availability of essential amino acids by fish, in general, is lower in canola meal than in soybean meal. The IAFFD database shows that the digestibility of essential amino acids contained in canola is on average about 7% lower than that of soybean meal.

The carbohydrate portion of canola meals is the most problematic from a formulation standpoint. In addition, the levels of indigestible carbohydrates, not including fiber, represent a substantial portion of the nitrogen free extract. These inherent nutrient characteristics are responsible for the relatively low digestible and metabolizable energy contents for fish.

Anti-Nutritional Factors

All rapeseed varieties contain glucosinolates. Enzymatic hydrolysis of these compounds during the process of digestion causes the release of isothiocyanates and goitrin. These function as anti-thyroid agents by inhibiting uptake of iodine by the thyroid gland. Additional iodine supplementation in the diet can compensate for the effects of thiocyanate ions. However, the effects of goitrin cannot be reversed with dietary iodine (Tookey et al., 1980).

Glucosinolates in canola varieties of rapeseed are considerably lower than traditional rapeseed, which ranges from 3 to 8 percent. Yurkowski et al. (1978) showed that feeding rainbow trout with traditional rapeseed caused thyroid hyperplasia and reduced plasma thyroxine concentration. Heat treatment of the meal inactivated the enzyme myrosinase, which hydrolyzes glucosinolates, but did not eliminate the glucosinolate content or improve performance of test diets containing rapeseed.

Table 8. Comparison of the composition and true availability of essential amino acids in canola and soybean meals

Essential Amino Acids	Canola Meal(1) Composition (3) (%)	Canola Meal (1) Availability (4) (%)	Soybean Meal (2) Composition (3) %	Soybean Meal (2) Availability (5) %
Arginine	6.1	83.6	7.4	96.9
Histidine	2.9	85.4	2.7	95.9
Isoleucine	4.0	80.3	5.4	94.2
Leucine	6.8	76.4	7.8	93.8
Lysine	5.3	81.2	4.6	96.1
Methionine	2.0	84.1	1.4	96.7
Cystine	2.6	--	1.5	92.5
Phenylalanine	3.9	81.0	5.6	94.8
Tyrosine	2.6	--	2.6	95.9
Threonine	3.9	89.1	4.1	95.8
Tryptophan	1.2	--	1.4	--
Valine	5.1	77.4	5.6	97.0

(1) Canola, seeds, meal solvent extracted. IFN 5 - 03 - 871. NRC, 2011.
(2) Soybean, seeds without hulls, meal solvent extracted. IFN 5 - 04 - 612. NRC, 2011.
(3) Expressed as percentage of protein, data adapted from National Research Council, 2011.
(4) Determined using rainbow trout (Higgs et al, 1994).
(5) Determined using rainbow trout (Yamamoto et al, 1998).

Another anti-nutritional component of rapeseed is erucic acid. This is a 22-carbon monounsaturated fatty acid. It has been shown to cause histopathological changes in skin, gill, kidney, and heart tissue of fish. However, the low erucic acid contents of canola varieties of rapeseed, along with low lipid contents in solvent extracted meals, virtually eliminates any anti-nutritional effects from the oil component of these meals. In fact, the NRC (1993) reported that no erucic acid pathologies have been associated with the inclusion of canola meals in practical diets for fish.

Formulation Recommendations

Ideally, rapeseed meals should never be used in feeds for aquaculture species. Only the meals made from canola varieties, with glucosinolate levels less than 30 μmoles and erucic acid levels less than 2% in the oil, have been shown to perform well in fish feeds.

Canola meals that have been processed by the prepressed solvent extraction method are the best choice for use in feeds because of the relatively higher protein and lower fiber contents. Even so, with a fiber content of over 9% and low available lysine and methionine/cystine levels, the economical limits of canola meals in fish feeds are usually less than 15%. It is also recommended to refrain from using canola meal in diets for small fish.

Peanut Meal

Peanuts, also known as ground nuts, can be a good source of protein and energy in fish feeds. The most commonly available meals are obtained as byproducts from the removal of high-quality oil. Peanut meals tested in diets for warm water species of fish seem to be highly palatable and exhibit excellent protein digestibility. In spite of these positive characteristics, their use in fish feeds is limited because of low lysine and methionine contents, and also because of regionally limited supplies.

Meal Products

The two most common meal products result from either mechanical or solvent extraction of the oil from whole peanuts without hulls. Table 9 presents both the proximate and amino acid composition of these meals and a comparison to the nutrient composition of dehulled, solvent extracted soybean meal.

Nutrient Composition

Both the mechanical and solvent extracted meal products contain about 48% protein. The mechanical extraction process, however, is not as efficient at removing oil. Consequently, the fat level is much higher in meal produced by this method than in solvent extracted meal. The difference is made up with a higher fiber level in solvent extracted meal.

Protein digestibility, as well as true amino acid availability, as measured in channel catfish, is excellent. The digestibility of amino acids in the IAFFD (2020) database of solvent extracted peanut meal determined for fish, in general, is approximately 1% below that of solvent extracted, dehulled soybean meal. However peanut protein is low in methionine and extremely low in lysine.

Anti-Nutritional Factors

Heat treated meals have no reported anti-nutritional properties that affect fish, though caution should be exercised in their use. Like cottonseed, peanuts have a high susceptibility to contamination with the fungus, *Aspergillis flavis*, which produced aflatoxin.

Formulation Recommendations

Both mechanically extracted and solvent extracted meals can be good and economical sources of protein and energy in fish feeds, under certain circumstances. Research conducted with catfish, *Ictalurus punctatus* (Robinson and Wilson, 1985), and tilapia, *Oreochromis mossambicus* (Jackson et al, 1982), indicates that use of these meals is limited by low levels of lysine and methionine. They are therefore most economical in diets that contain fishmeal and/or blood meal, which are high in lysine. Hertrampf and Piedad-Pascual (2000) recommend the following maximum levels of mechanically or solvent extracted peanut meals (without hulls) in diets:

Herbivorous/omnivorous fish	15%
Carnivorous fish	10%
Crustaceans	8%

Sunflower Meal

Sunflower (*Helianhus annua*) is an oilseed crop that is grown in many areas of the world because of the high food value of its oil and the ability of the plants to adapt to a variety of climates and soil conditions. The whole seed has a high oil content, ranging from 25% to 32%, which seems to be dependent on growing conditions. Protein and fiber levels are about 16% and 28%, respectively. Sunflower meals are produced from the seed, following oil extraction. While research on the use of these meals in fish feeds has been limited, published studies with rainbow trout (Tacon et al., 1984) and tilapia (Jackson et al., 1982) have shown them to be a good source of protein, though low in lysine. The major impediment to their use is the relatively high levels of fiber.

Table 9. Nutrient composition of commonly available peanut meals compared to dehulled, solvent extracted soybean meal

Description Internat. Feed No.	Peanut, Meal mech. extd. (1) 5 – 03 - 649	Peanut, Meal solv. extd. (2) 5 – 03 - 650	Soybean meal, w/o hulls, solv. extd (2) 5 – 04 - 612
Moisture (%)	7.0	8.0	10.0
Crude Protein (%)	48.1	49.0	48.5
Crude Fiber (%)	6.9	9.9	0.9
Ether Extract (%)	5.8	1.3	3.4
Ash (%)	5.1	5.9	5.8
Arginine (3)	10.5	6.9	7.4
Histidine (3)	2.2	2.0	2.7
Isoleucine (3)	3.5	2.9	5.4
Leucine (3)	6.3	6.1	7.8
Lysine (3)	3.1	5.4	4.6
Methionine (3)	1.0	1.3	1.4
Cystine (3)	1.5	1.0	1.5
Phenylalanine (3)	4.9	3.5	5.6
Tyrosine (3)	3.4	2.2	2.6
Threonine (3)	2.6	3.4	4.1
Tryptophan (3)	1.0	0.6	1.4
Valine (3)	4.3	5.0	5.6

(1) Adapted from National Research Council, 1982
(2) Adapted from National Research Council, 2011
(3) Expressed as percentage of protein, data adapted from National Research Council, 2011.

Meal Products

The best choices for sunflower meals for use in aquaculture feeds are those that are produced from decorticated seed. By removing most of the seed hulls before processing, meals that are low in fiber and higher in protein can be produced with either the expeller or solvent methods of oil extraction. Table 10 presents both the proximate and amino acid composition of these meals and a comparison to the nutrient composition of dehulled, solvent extracted soybean meal.

Nutrient Composition

Proximate composition of the meals varies slightly according to the variety of seed, but more with the method of processing (Table 10). Expeller processed meals contain more fat and fiber and lower quantities of protein than do meals produced by solvent extraction. Both meals have higher concentrations of the sulfur containing amino acids, methionine, than does solvent extracted soybean meal without hulls.

The temperature involved in the process of oil extraction also influences the quality of protein in the meal. Solvent extraction at relatively low temperatures reduces the destruction and/or loss of lysine, while dry heating at high temperatures causes reduction in lysine content and availability (Renner et al, 1953).

Table 10. Nutrient composition of commonly available sunflower meals compared to dehulled, solvent extracted soybean meal

Description Internat. Feed No.	Sunflower, Meal mech. extd., w/o hulls (1) 5 – 04 – 738	Sunflower, Meal solv. extd., without hulls (2) 5 – 04 – 739	Soybean meal, w/o hulls, solv. extd (2) 5 – 04 - 612
Moisture (%)	7.0	7.0	10.0
Crude Protein (%)	41.4	46.5	48.5
Crude Fiber (%)	12.2	13.0	0.9
Ether Extract (%)	8.0	0.5	3.4
Ash (%)	6.6	7.1	5.8
Arginine (3)	8.3	7.5	7.4
Histidine (3)	2.2	6.1	2.7
Isoleucine (3)	3.54.3	4.5	5.4
Leucine (3)	6.0	5.6	7.8
Lysine (3)	3.9	3.7	4.6
Methionine (3)	2.3	3.2	1.4
Cystine (3)	1.6	1.5	1.5
Phenylalanine (3)	4.3	2.6	5.6
Tyrosine (3)	2.4	1.6	2.6
Threonine (3)	3.3	3.2	4.1
Tryptophan (3)	1.6	0.8	1.4
Valine (3)	4.8	4.9	5.6

(1) Adapted from National Research Council, 1982
(2) Adapted from National Research Council, 2011
(3) Amino acids are expressed as percentage of protein

The most notable part of the nutrient composition of sunflower meals is the high content of fiber. Meals obtained from whole seeds, without the hulls removed, can contain up to 32% crude fiber (National Research Council, 1982). Improvements in oil extraction and meal processing have lowered the crude fiber and ether extract levels. However, even solvent extracted meal from decorticated seed contains 11 to 12% crude fiber.

Anti-Nutritional Factors

Tacon (1984) reported that sunflower meals contain a variety of endogenous anti-nutritional factors. One of these, chlorogenic acid, is reported to function as an effective trypsin inhibitor (Kanto, 1988). It is thought that part of the reason for improvement in nutritive value of sunflower meal by mild heating may be due to the destruction of this compound.

Formulation Recommendations

Reports from the limited amount of research on sunflower meals in diets for fish suggest that they can be a good source of protein and energy. Apparently, fish readily consume diets with rather high levels of sunflower meals, and there are no major problems with anti-nutritional components when properly processed. However, the relatively high fiber contents and low level of lysine will necessarily limit use in high performance feeds.

Maximum dietary levels of sunflower meal between 10 and 15%, depending on fiber content of the meal and contributions of fiber from other diet components, are appropriate in high quality fish feeds. It would also be advisable to refrain from using sunflower meals in brood-fish and crustacean diets until published information on performance is available.

Final Comment

In concluding, it is necessary to comment on the use of synthetic amino acids in formulating diets that primarily contain ingredients of plant origin. Without exception, all of the plant-protein supplements are lower in lysine than fishmeal, any many are also lower in total sulfur-containing amino acids. Use of purified amino acids is one obvious way to compensate for deficiencies resulting from the presence of these meals in a diet (NRC, 2011). However, presently available research literature is unclear on the effectiveness of supplementing fish diets with purified, single amino acids. There are studies that indicate efficient utilization of crystalline amino acids by fish and other studies that point to the contrary conclusion (NRC, 2011).

The reason for the differences in results is attributed to a metabolic dyssynchrony between the absorption rates of crystalline amino acids and those in intact protein. It is generally accepted that multiple feeding frequencies lead to better metabolic utilization of crystalline amino acids. Until this issue is more fully investigated, it is recommended that crystalline amino acids be used conservatively in feeds that will not be fed in multiple daily feeding programs.

About the Authors

Tim O'Keefe is an aquafeed specialist who has over 45 years of experience in fish and shrimp nutrition, diet formulation, and feed manufacturing.

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Tim received his formal education and training at Texas A & M University where he received a Bachelor of Science degree, majoring in biochemistry, and a Master of Science degree in fisheries sciences. At the beginning of his career he spent a few years working for vertically integrated farming companies that produced salmon and trout in the northwestern part of the United States and in British Columbia, Canada. Before founding Aqua-Food Technologies, he worked for 17 years as the Aquaculture Feed Division manager at Rangen, Inc., a diversified agriculture company with headquarters in Idaho. He managed both the domestic and international operations of the company's aquaculture feed division.

Tim started Aqua-Food Technologies, Inc. in 1998. During the ensuing 22 years, his company contracted every year with the American Soybean Association and US Soybean Export Council to provide technical feed support. AFT also contracted with numerous international feed companies to provide technical assistance with the manufacturing of a wide variety of feeds for fish and shrimp. He and his partner, Mark Newman, worked with clients in Bangladesh, Brazil, Egypt, Honduras, India, Indonesia, Malaysia, Mauritius, Mexico, Myanmar, Nepal, Pakistan, People's Republic of China, the Republic of Philippines, the United States, Vietnam, and Zimbabwe.

Mark Newman has worked for more than 40 years in the fields of aquaculture, aquaculture nutrition, the manufacture of aquafeeds, and fisheries biology.

He worked for a major U.S. feed manufacturer in the U.S. and Ecuador for 20 years in the areas of nutrition, quality assurance, management, and technical support. He has commercially farmed marine shrimp and has conducted nutritional research with tilapia, Pacific salmon, freshwater prawns, a variety of marine fish, and marine shrimp. Presently he is an aquaculture nutrition and feedmill consultant providing technical assistance to feedmill companies in many parts of the world, including: Latin America, South East Asia, Pakistan, Bangladesh, India, and Egypt.

Soy In Aquaculture Program

This technical paper was created through the USSEC Soy In Aquaculture (SIA) program and the USSEC Southeast Asian Regional Program. USSEC works with target audiences in Southeast Asia and globally to show the utility and benefits of using United States soybean products in aquaculture diets.

The SIA program replaces the Managed Aquaculture Marketing and Research Program (the AquaSoy Initiative, funded and supported by the United Soybean Board and American Soybean Association) which was designed to remove the barrier to soybean meal use in diets fed to aquaculture species.

The objective of the SIA is to optimize soy product use in aquaculture diets and to create a preference for U.S. soy products in particular, including but not limited to U.S. soybean meal, soybean oil, soybean lecithin, and "advanced soy proteins" such as fermented soy and soybean protein concentrate.

This paper follows the tradition of USSEC to provide useful technical materials to target audiences in the aquaculture industry.

For more information on soybean use in aquaculture and to view additional technical papers, please visit the Soy-In-Aquaculture website at www.soyaqua.org.

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