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Review Article

Unlocking the Hidden Potential of Plant Proteins in Fish Nutrition Using Technology

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ABSTRACT

Plant sources of protein are the most appropriate alternative for Fish Meal (FM) for preparation of fish diet. Therefore, plant protein is the lower price, greater availability and improved consistency of composition. Compared to fish meal, they vary in their nutrition and biological values and lower palatability. In order to improve nutritional quality of plant protein sources by using processing technologies such as Solid state fermentation has been considered for the production of enzymes, antibiotics, extrusion cooking, other value-added products to produce high quality aquatic feeds. Besides that, biotechnology to get better safety, freshness, color, flavor, texture, taste, nutritional characteristics and shelf-life of cultured food products. In addition, mixed feeding from plant protein source in which high protein diet is alternated by a low protein diet could result in improved nutrient utilization. Thus, to take practical and possible measure in order to enhance nutritional quality of plant protein sources by using extrusion cooking processing, genetically modified, biotechnology, feed additives, nano-particles and solid state fermentation technologies. Hence, the issue of cheaper sources of plant protein feed ingredients would be requisite to fully replace fish meal for the improvement and sustainable of fishery industry in developing countries.

Keywords: Biotechnology; Feed additives; Plant protein; Solid state fermentation

INTRODUCTION

Fish production is expected to increase further to meet the demand of an increasing population in the world; this will need production of more fish feed. The dependence on addition of the plant-protein constituents in fish feeds has increased because of its low cost and presence of balanced amino acids [1]. Different plant sources such as oil seed meals, maize, bagasse mix are available, which are produced as byproducts from various industries [2]. Besides that, plant protein supplements, cereal grains and grain by-products are extensively used in feeds for aquaculture species. Moreover, laguminous crops include peas and beans of which soybean has used plant protein source in aqua feeds [3].These sources are easily available at low or no cost. Various scientists have evaluated an efficiency and use of these sources as a partial or complete alternate of FM in fish diets [1,2].

Proteins compose approximately 70% dry weight of the organic material in fish tissue; therefore, protein content is one of the main nutritional compounds of fish feeds. The replacement of fish meal with cheaper ingredients of plant origin in fish feed is necessary because of rising cost and uncertain availability of fish meal [4]. Several plant protein sources can be used to partially or almost totally replace dietary fish meal [5], supplied that the essential amino acid requirements of the fish species are met, the palatability of the diet is improved and the levels of Anti-Nutritional Factors (ANFs) are reduced [6].

Feed of plant origin has a big potential for fish feed industry and they are a cheaper source of high quality proteins as compared to animal sources. Fish meal and plant protein sources are very different in the amount of proteins, structure of amino acids, energy availability and amount of mineral matter [7]. Lower price, greater availability and improved consistency of composition are the significance of plant protein sources (Table 1). Compared to fish meal, though they vary in their nutrition and biological values and generally have lower palatability. Deficiencies of Essential Amino Acid (EAA) often cause decreases in growth performance and feed efficiency ratios [8]. Locations, seasonal changes, growth condition, agricultural practices as well as variations between individual plants can affect the nutritional composition of plant materials [9,10]. Another limiting factor to using plant derived proteins in the existence of ANF toxicants that present as protease inhibitors cyanogens, lectins, saponins, alkaloids, phytic acid, tannins, phytoestrogens and glucosinolates [11]. ANF can adversely affect digestion, absorption and physiological utilization of protein and amino acids and can limit the palatability and the nutritive utilization of protein [11-20].

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Table 1: Essential Amino Acid (EAA) profiles of various plant protein sources (%).

Plant protein sources	Arg	His	Lle	Leu	Lys	Met	Phe	Thr	Trp	Val
SBM	3.2-4.2	1.2-1.4	2.0-2.3	3.7-4.4	2.8-4.0	0.5-0.9	2.3-2.4	1.7-2.4	0.5-0.6	2.2-2.5
SPC	4.5-6.4	1.5-2.0	2.6-3.7	4.7-5.9	2.8-4.7	0.9-1.0	3.0-3.6	2.5-3.1	0.9-1.0	2.7-3.8
SPI	6.7-7.5	2.2-2.4	3.8-4.6	6.8-7.2	4.5-5.7	1.1-1.3	4.5-4.7	3.1-3.6	1.1	4.0-4.6
CGM	1.9-2.2	1.1-1.4	2.0-2.8	9.7-11	1.0-2.1	0.9-1.8	3.3-4.4	1.7-2.3	0.3-0.5	2.4-3.4
CGF	0.9-1.2	0.6-0.7	0.6-0.7	1.6-2.2	0.6-0.7	0.3-0.5	0.7-0.8	0.7-0.8	0.1-0.2	0.9-1.1
DDGS	0.9-2.2	0.6-1.0	0.9-1.5	2.4-4.0	0.5-1.1	0.5-0.8	1.3-1.7	0.8-1.3	0.2-0.3	1.3-1.8
CSM	0.9-5.7	0.5-1.4	1.2-2.4	1.0-2.8	1.0-2.2	0.5-1.1	1.6-3.3	0.9-1.7	0.3-0.5	0.9-1.3
Alfaalfa meal	0.6-1.1	0.3-0.5	0.7-1.1	1.2-1.6	0.6-1.0	0.3-0.4	0.7-1.1	0.6-0.9	0.3-0.5	0.9-1.3
RSM/Canola	1.1-2.5	0.5-1.2	0.8-1.8	1.2-2.9	1.2-2.5	0.4-0.8	0.7-1.6	0.9-1.8	0.3-0.5	1.0-2.2
Lupin meal	3.8-4.4	0.9-1.2	1.4-1.7	2.3-2.8	1.4-2.1	0.2-0.4	1.3-1.4	1.3-1.5	0.3	1.3-1.6
Pea meal	1.5-2.5	0.5-0.6	0.9-1.1	1.5-1.7	1.5-1.7	0.2-0.3	1.0-1.1	0.8-0.9	0.2	1.0-1.2
Lentil meal	2.3	0.9	1.1	2.1	1.6-1.9	0.2	1.4	0.9	0.2	1.4
Chickpea meal	1.5-2.0	0.2-1.1	0.8-1.1	1.1-2.2	1.4-1.8	0.2-1.0	1.1-1.5	0.7-1.0	0.2	0.8-1.3
Navy meal	6.9	7.1	2.1	6.5	6	1.1	3.9	3.4	NA	3.3
Pinto meal	6.4	2.8	4.4	8.1	6.9	1.1	5.4	4	0.9	5
Black meal	5.5-6.7	1.9-2.9	4.1-4.3	7.7-8.6	6.7-7.1	2.0-2.7	5.1-5.8	3.1-4.4	1.4-1.5	4.9-5.1

Note: All are on a dry matter basis; SBM: Soyabean Meal; SPC: Soy protein Concentarate; SPI: Soy protein Isolate; CGM: Corn Gluten Meal; CGF: Corn Gluten Feed; DDGS: Distillers Dried Grains with Solubles; CSM: Cottenseed Meal; RSM: Rapeseed Meal

LITERATURE REVIEW

Processing technologies used to enhance nutritional quality of plant protein sources

Solid State Fermentation (SSF): It is a fermentation process in which microorganisms are grown on solid substrate without the presence of free liquid [21] and it has been intensively for the production of enzymes, antibiotics, surfactants and other value-added products [22]. As compared to the submerged fermentation in which nutrients are present in dissolved form in a large amount of water, SSF a great commercial potential due to its lower wastewater production and operating expenses, simpler fermentation medium requirement, superior productivity and easier prevention of bacterial contamination [23]. Agro-industrial residues are sugar cane bagasse, cereal straws, brewer's spent grain and corn stover and they have been used as substrates for SSF to maximize their utilizations and to address the waste disposal issues [24]. Thus, SSF may have a great potential in producing enzymes to break down its fiber to improve its digestibility [25].

Enzyme complex solid state fermentation: Addition of enzymes in fish diets an alternative to improve the utilization of nutrients coming from plant ingredients. In addition to reducing production costs by increasing digestibility, it reduces the load of pollutants from waste, enabling better sustainability [26]. The greatest benefit of this additive is its combination of natural enzymes (protease, β-amylase, cellulase, xylanase, α-galactosidase, pectinase, phytase, endoglucanase, sucrase, and others), which can improve the digestibility of plant feedstuffs in the diet [26].

Feed additives and related technologies: Plant protein is important potential for addressing the problem of phosphorus pollution, since plants do not contain the high levels of phosphorus found in animal protein. Scientific research in the area is focusing on the investigation of various plant species and plant-animal protein mixes, as new sources for protein for aqua feeds for shrimp [27]; mollusks [28] and finfish [29]. In addition, brewer's yeast is another protein source for finfish [30], along with plant lipid substitutes for fish oils. One of the difficulties in using plant proteins in aqua feeds is the need for appropriate processing to destroy anti-nutritional compounds which may harm the fish once fed. Biotechnology also improves safety, freshness, color, flavor, texture, taste, nutritional characteristics and shelf-life of cultured food products. Tools are already under development, or commercially available, that can detect and assay toxins, contaminants and residues in aquatic products [31,32]

Extrusion technology: According to Reddy extrusion cooking is one of the most promising methods to produce high quality aquatic feeds. Production of aquatic feeds fell into one of four production categories: floating feeds, sinking feeds, slow sinking feeds and soft moist feeds [33]. Extruders are also capable of producing sinking floating and soft moist foods. The ability to control the density of a finished product allows the production of floating or sinking aquatic feeds that are tailored for particular species of aquatic animals. Floating aquatic feeds are in the density range of 320 to 400 g/1 and expanded pellets varying in diameter from 1.5 to 10 mm. typical floating feeds are extrusion cooked at the high temperature and lower moisture levels and expands upon exiting the diet approximately 125 to 150% of the original whole size [34]. Sinking aquatic feeds are typically in the density range of 400 to 600 g/l. These products are generally 1.5 to 4.0 mm in diameter and predominantly are used for shrimp feeding. Slow sinking aquatic feeds are in the density range of 390 to 410 g/1. These products sink in salt water. These types of feeds are being used in the salmon raising industry. The technology of producing soft moist feeds is very similar to producing floating aqua feeds on the extruder [34]. Aqua feeds can be manufactured through extrusion processing that hit precise targets in terms of size, durability, density and nutritional composition at a competitive price. Twin-screw extrusion technology can be used to process ultra-high fat aqua feeds with fat levels above 17% [35]. Extrusion of soy products has to improve digestibility and growth performance when included in carnivorous [36] or omnivorous [37] fish diets. Extrusion cooking has been found to improve carbohydrate digestibility through gelatinization of starch and fiber [36]. The conditions of the extruder (e.g., temperature, moisture, screw speed, retention time) can be manipulated to cause desirable physical and chemical changes within the extrudate [38]. This allows a greater susceptibility to enzymatic hydrolysis [39] either by gastrointestinal enzymes, or through fermentation.

Role of plant protein ingredients and additives

Organic acids and their salts: Sodium, potassium or calcium appear to have the potential to improve growth performance of some animals [40]. The positive effects of organic acids and acid salts on animal growth performance and health are exhibited *via* three different mechanisms in Table 2 feed, digestive tract and metabolism [41].

During feed storage, there may be a certain level of contamination with fungi, bacteria and yeasts. The lower pH inhibits microbe growth and metabolism, thus reducing the risk of contamination from pathogenic organisms and their metabolites.

Hormone: Growth Hormone (GH) is one of the polypeptide hormones secreted by the somatotrophs in the anterior portion of the vertebrate pituitary gland. GH is involved in the regulation of somatic growth and the maintenance of protein, lipid, carbohydrate and mineral metabolism [42]. Zohar found that GH may be the most promising agent for growth promotion in aquaculture [43]. On the other hand, the accessibility of natural GH is limited because its preparation from fish pituitary glands is not economical. The complementary DNA (cDNA) encoding fish GH has been sequenced and the recombinant GH (rGH) has also been shown to be potent in enhancing the growth rate of fish by injection or by immersion [44,45]. According to Tsai et al., demonstrate that the growth of juvenile black seabream (*A. srhlegeli*) could be enhanced by using renatured fish rGH produced by *E. coli* as a feedstuff additive [45].

Enzymes: According to Phadke et al., stated that enzymes are basically types of protein in biological systems [46]. They are commonly used as catalysts in order to catalyze the rate of reaction. There are different kinds of enzymes which include phytase, xylanase, cellulase, lipase, protease, amylase and many more which can increase the nutrient availability, nutrient absorption during digestion, increase the rate of fish growth and assist survival of fish in early stages of life (Table 3). In addition, it makes the feeds more economical. Enzyme application may give a solution of high larval mortality of aquatic animals. Feeding larvae with enzymes would be beneficial [46]. It has been stated the enzymes are also used in the feeds based on plant-derived raw materials in various countries in order to increase digestibility and a better weight gain and feed conversion [47].

Among the main enzymes used in the mixed feed fisheries industry are phytase, carbohydrase, protease and lipase [48]. When it is considered that feed cost takes place in the largest cost of the aquaculture industry, economic feed production is inevitable and economic feed production will also be possible with the use of inexpensive vegetable protein source in feeds. The addition of the enzymes which facilitate digestion of vegetable protein sources to feeds [48,49]. Ebru et al., suggested that when animals are under restricted feeding regimes as the beneficial effects of enzyme addition may be hidden by increased feed intake under an ad libitum feeding regime [48].

Immune-stimulants in aqua feeds: Immuno-stimulants have been used as feed additives for several years in aquaculture, and yeast β-glucan may be the one with the longest track record. In nature, β-glucans are widespread and have been characterized in microorganisms, algae, fungi and plants [50]. According to Raa et al., immune-stimulants are being used in the aquaculture sector to reduce mortality due to infections and to improve general performance of animals or fish [51]. Immune-stimulants may provide particular benefits when used in order to: Reduce mortality due to opportunistic pathogens, prevent virus diseases, enhance disease resistance of farmed shrimp, reduce mortality of juvenile fish, enhance the efficacy of anti-microbial substances and enhance the resistance to parasites and enhance the efficacy of vaccines.

Table 2: Mechanisms of organic acids and their salts.

Site of action	Effects
Feed	pH reduction
	Antimicrobial effects
	Reduced buffering capacity
Digestive tract	pH reduction in stomach
	Increase in efficiency of pepsin (pH optimum 2.5 and 3.5)
	Antimicrobial effect
	Complexing agent (Ca^{2+}, Mg^{2+}, Fe^{2+}, Cu^{2+}, Zn^{2+})
	Antimicrobial effect
Metabolism	Energy source

Table 3: Types of enzyme and their function.

Enzyme	Function
Amylase	Utilization of starch and complex polysaccharides
Arabinase, pectinase	Degradation anti- nutritional factors, improves animal's energy level
Mannanase	Degradation of mannans, a Non-Starch Polysaccharide (NSP)
Cellulase	Degradation of cellulose in plant feedstuffs
Keratinase	Degradation of feather keratin for production of feather meal
Lipase	Emulsification and utilization of oils and fats
Phytase	Increasing bio availability of phosphorus, trace minerals and protein in feedstuffs
Protease	Utilization of animal and plant proteins
Tannase	Elimination of tannin
Xylanase	Hydrolysis of non-starch polysaccharides like pentosans
Galactosidase	Acts on glalactosides to reduce anti-nutritional factors
Glucosidase	Acts on glucans (NSP) to unlock nutrients
Invertase	Converts sucrose to glucose and fructose and improves animal's energy level

Nano-technology materials: Feed additives in nano forms have different effects from enhancing growth and immunity through antioxidant effect to their use in fewer amounts than its bulk counterparts which enhances ration criteria [52, 53].

Different metal and metal oxide nano particles are screened for their antimicrobial activities against a wide range of bacterial and fungal agents including certain freshwater cyanobacteria [54]. Among different nanoparticles, synthesized copper oxide (CuO), zinc oxide (ZnO), silver (Ag) and silver doped titanium dioxide (Ag-TiO_2) showed broad spectrum antimicrobial activity [54]. Since CuO, ZnO and Ag nano particles have higher antimicrobial activity; they may be explored for aquaculture use. Zinc oxide nano-particles as one of metal oxides are versatile because they enter in a wide variety of applications ranging from sensing, catalysis, energy storage, electronic devices and biomedical applications. Chemically, zinc oxide (ZnO) and nano-zinc oxide (nZnO) have the same chemical formula which suggests similar Zinc to oxygen ratio, but at the nano scale atoms are arranged with a wider energy level confinement and smaller size Zn, that could lead to more reactive atoms as the surface is increased [55]. The dietary supplementation of Zn, nano Zn and Cu have produced better survival and growth in *M. rosenbergii* PL [56]. Furthermore, iron-based nanoparticles are also used for soil and groundwater remediation and water treatment processes [57].

Probiotics: It is a live microbial feed supplement which beneficially affects the host animal by improving its intestinal balance and growth promoting factors produced by microorganism. Moreover, probiotics have been investigated as a potential dietary supplement that can positively contribute to an individual's health [58,59]. Probiotic strains commonly used for aquaculture practices include members of the *Lactobacillus, Lactococcus, Leuconostoc, Enterococcus, Carnobacterium, Shewa nella, Bacillus, Aeromonas, Vibrio, Enterobacter, Pseudomonas, Clostridium*, and *Saccharomyces* genera has stated that carps fed with the feeds added brewer's yeast S. cervisiae [60]. According to El-Haroun et al., have stated that after adding Biogen (commercial probiotic) into Egyptian tilapia (*O. niloticus*) fingerling feeds in various rates, probiotic added feeds show higher growth performance and benefit when compared to the feeds in which probiotic is not added [61]. It is effect of probiotic Bacillus on the feed digestion of rainbow trout (*O. mykiss*) tiddlers, growth and survival rate [62]. Adding appropriate density of commercial probiotic Bacillus into the initial feed of trout is beneficial especially for its growth and survival in the growing conditions [63] state that adding probiotics into the pacific oyster (*C. gigas*) larval culture enhances the development of oyster larvae completely.

Prebiotics: It is like organic acids are mainly used in order to sanitize the feed containing various infectious and pathogenic agents [64,65]. They are non-digestible feed ingredients that beneficially affect the host by selectively stimulating the growth or activity of one or a limited number of bacterial species, already resident in the gut and thus attempt to improve host health [65,66]. Moreover, prebiotics in fish and shellfish have explore the following parameters: effect on growth, feed conversion, gut microbiota, cell damage/morphology, resistance against pathogenic bacteria and innate immune parameters such as Alternative Complement Activity (ACH50), lysozyme activity, natural haemagglutination activity, respiratory burst, superoxide dismutase activity and phagocytic activity [65-67].

DISCUSSION

Genetic modification plant protein sources

Genetically Modified (GM) plants are plants whose genetic material is modified in a way which is not found in nature under natural conditions of crossbreed or natural recombination [68,69]. Irrespective of the technology used, integration of the novel DNA happens through so called illegitimate recombination (non-homologous end-joining) rather than homologous, or site-directed, recombination [70]. According to Sissener et al., the use of genetically Modified (GM) plant proteins in fish diets on the use of Round-up Ready Soybeans (RRS) and Bt-maize in salmon diets [69]. These show varying results, the major trend being that no or very small effects on fish growth or feed utilization. Few alterations in organ sizes are found, except for the gastrointestinal tract, which seems to be affected by the plant ingredients especially, but regardless whether the plant is GM or not [71-75].

CONCLUSION

Replacement of fish oil and fishmeal by lipid and protein from vegetable sources is a necessity to make the aquaculture industry sustainable. However, fish oil replacement may affect the product quality and health benefits for the consumer, as the fish fillet will contain less of the marine long-chained polyunsaturated omega-3 fatty acids. Plants modified by introducing genes from microalgae to produce omega-3 fatty acids are under development, and commercial production. Amino acid composition of plant proteins is another way in which plants can be modified to be more attractive as fish feed ingredients. Maize with increased level of lysine is already on the market (LY038) and has been tested with positive results in broilers. Corn gluten is a promising protein source for use in fish, but low lysine level is considered a limiting factor.

CONFLICT OF INTEREST STATEMENT

The authors declare that there are no conflicts of interest. There is no funding source available.

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