A review on alternative plant protein sources available for future sustainable aqua feed production

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Abstract
Global crisis of fish meal for aqua feed production imposed the researchers to search an alternate non-conventional source of protein. Till date, most of the research conducted on replacement of fishmeal, used plant based protein ingredients. Plant based feed ingredients are rich in protein, but contain high amount of anti-nutritional factors which curb their usage in fish diet. In order to get rid of the anti-nutritional factors, present in the plant based feed ingredients, generally fermentation, solvent extraction, heat treatment and protein isolation methods are used. Among the reviewed plant based protein sources, soybean meal has the highest protein content but ultimate dependence on soybean meal has its own disadvantages. Rubber seed meal (26% protein) could replace 50% of fish meal in carp diet whereas canola meal (37% protein) and Jatropha kernel meal (27.2% protein) have a good amino acid profile, makes them suitable for the incorporation in fish diet. Palm kernel meal (16.6% protein) could not replace a considerable amount of fish meal in the aqua feed due to its low protein content. However, peanut meal (48%) could significantly reduce the inclusion of fish meal in the diet with proper lysine and methionine supplementation. In this paper, research works published on fish meal replacement by using plant protein sources (soybean meal, rubber seed meal, canola meal, palm kernel meal, jatropha kernel meal, peanut meal) were reviewed.

Keywords: Aqua feed, protein source, fish meal, plant based feed ingredients, anti-nutritional factors

Introduction
The rapid expansion of human population, in the past centuries, created nutritional related issues, especially food security concerns throughout the world. In order to ensure food security and to maintain an uninterrupted supply of nutritious food, supply of fish, the high proteinaceous cheapest food – needs to be increased. However, on the other side, harvesting of fish from natural resources had declined sharply, in the recent decades, which directly paved the way for aquaculture sector. The annual growth of global fish consumption increased twice as population growth, showing that, in future, aquaculture sector will play a very crucial role in implementing the goal of FAO i.e. “A World without Hunger and Malnutrition.” (FAO, 2018) [55].

Aquaculture represents 47% of total global fish production and it has been projected to increase its share (52%) in 2025 (SOFIA, 2018) [55]. In aquaculture production, feed is the single largest input. So the expansion of aquaculture practices – to achieve the projected fish production – needs to be supported by the expansion of aqua-feed industry. Traditionally, for the aqua feed production fishmeal is considered as a main dietary protein source. However, in the recent past, decreased supply and high demand for fish meal led to its price hike and total dependence of fish meal is not advisable (Kaushik et al., 1995; Fournier et al., 2004). Furthermore, the gradual increase in the price and restricted availability of the fish meal convinced researchers and feed manufacturers to search an alternate source of protein for sustainable aqua feed production.

In this context, plant protein sources are easily available and generally fetch a lower market price compared to fish meal. In spite of being rich in protein, the presence of anti nutritional factors limits their inclusion in the aqua feed. Anti-nutritional factors are known to adversely affect the digestive as well as the metabolic activity of the cultured species. Among the anti nutritional factors, phytic acid and trypsin inhibitors cause major issues. Hence detoxification has to be done before incorporating the plant protein sources in fish feed. Among the plant protein sources, some of the ingredients such as soybean meal, are already in use, as a
replacement of fish meal and some of the ingredients such as corn gluten meal, palm kernel meal, cottonseed meal, canola meal, jatropha kernel meal and peanut meal have high possibilities for future incorporation in the fish diet, as a partial or complete replacement of traditional protein sources. The present review paper briefly discuss about the possible alternative plant based protein ingredients available for future aqua feed production.

**Soybean meal**

Soybean meal (SBM) is considered as one of the most nutritive feed ingredients commonly used in animal feed industry. Proximate composition of soybean meal is shown in Table no 1. Due to its balanced amino acid profile, it can be used to replace the animal protein in the fish diet. Anti-nutritional factors present in SBM, such as trypsin inhibitor, saponins, lectins, and oligosaccharides, have a negative impact on nutrient digestibility and growth performance of fish. Mambriini et al. (1999) reported that 75% supplementation of dietary protein using soy protein concentrate as a fish meal replacement showed better amino acids digestibility than pure fish meal diet. According to Liu et al. (2000) 8% inclusion of soy protein concentrate could replace 22% of fish meal in *Peneaus monodon* diet. It is reported that 40-50% of the animal protein could be replaced by 25-30% of SBM in shrimp feed without compromising the growth (Swick et al., 1995). Soy protein concentrate (Soycomil) could replace 50% of fish meal in rainbow trout feed (Mambriini et al., 1999) [37]. Similarly, Stickney et al. (1996) reported, 50% inclusion of SPC as a fishmeal replacement in trout diet resulted in better growth performance. However, a higher rate of inclusion i.e @ 70 to 100% lead to the reduced growth rate of trout. After a 10 weeks experimental trial, Wilson & Poe (1985) concluded that incorporation of raw or insufficiently treated (heat treatment) soybean meal could cause growth reduction in channel catfish. He reported that reduced trypsin inhibitor could improve the growth performance of the fish. Decreased growth rate, might be due to the presence of heat stable anti-nutritional factors. According to Escaffre et al. (1997) SPC could be incorporated, up to 40% in the carp diet, without compromising the survival and growth of carp larvae. It has been found that price of the soybean meal as an alternative protein source is rising gradually and creates conflict between animal and human food sector (Kaushik et al., 1995) [24]. Hence total dependence on soybean meal as a fish meal replacement will not serve the purpose.

**Rubber seed meal**

The main product of rubber tree are the latex and seed (Alenius et al., 1995) [4]. Rubber seed consist of kernel (65%) and shell (35%) (Joseph et al., 2004) [23]. Rubber seed is the most promising nutritive feed ingredient which is having a crude protein level of 20 to 30% with a balanced amino acid profile. Proximate composition of rubber seed meal are shown in the Table 2. The main limitation of incorporating rubber seed meal, in the fish diet, is the presence of several anti-nutritional factors such asphytic acid, oxalic acid, hydrogen cyanide, saponin, tannin, and trypsin inhibitor (Aguirue et al., 2017) [3]. The cyanogens are the major anti nutritional factor present in rubber seed. It contributes to gastrointestinal issues and reduce metabolic activity when ingested (Thangaraj, 2016) [59]. However, processing of the rubber seed meal helps in reducing the anti-nutritional factors. Storage of the rubber seed meal for 2 months or more was proven to be an effective way of lowering the anti-nutritional factors, but this is not sufficient to lower the anti-nutritional factors as per the requirement of feed industries or nutritionists (Oluodo et al., 2018) [46]. Therefore, soaking, heat treatment, fermentation and enzymatic treatment are used as an alternative detoxification methods (Aguirue et al., 2017) [3]. Suprayudi et al. (2015) reported better growth response of carp juvenile fed with treated rubber seed meal with an inclusion level of 50%. On the other hand, Deng et al. (2017) reported that inclusion of rubber seed meal more than 30% in tilapia (*Oreochromis niloticus* x *O. aureus*) diet cause growth depression. The reason behind this contradictory result in the growth performance might be due to the variation in the residual anti nutritional factors. Similarly, Sharma et al. (2014) documented that *Laobe rohita* fingerlings fed with rubber seed meal @ 20% inclusion showed best without affecting the growth response. Fawole et al. (2016) reported that nutritional quality and digestibility of rubber seed meal could be improved by using protein isolation method.

**Canola meal**

Canola (*Brassica napus*) is primarily known for its oil content. However, it has good amount of protein and it also used as a feed ingredient in animal nutrition. Generally, the nutritional quality of canola meal depends on the intensity of heat imparted during oil extraction, residual oil content and the levels of anti-nutritional factors (glucosinolates) and their by-products. During the oil extraction process, excessive heating results in reduction of protein and amino acid digestibility, especially lysine. According to Friedman, (1996) amino acid profile of canola protein is the best among all the available vegetable protein sources. Proximate composition of canola meal is shown in Table 3. Mwachireya et al. (1999) reported that canola meal has high fibre content (10.6%) and it could be converted into protein concentrate following aqueous extraction method. Canola protein concentrate contains almost similar crude protein as fish meal and comparatively high lysine and methionine than soybean meal. Anti-nutritional factors such as heat liable glucosinolates and heat stable phytic acid, tannins and phenolic compounds are present in canola meal. Newkirk et al. (2003) reported, that total glucosinolate content of Iranian variety and Canadian variety canola meals are 5.74 μmol g⁻¹ and 7.2 μmol g⁻¹, respectively. Canola meal is commonly used in the diet of salmon, trout, carp, catfish, seabass tilapia, perch, sea bream, and shrimp (H.R. Enami, 2011) [12]. Hardy and Sullivan, (1983) suggested 20% inclusion of canola meal in the diet of rainbow trout (*Salmo gairdneri*) without affecting the growth. Similarly, Shafiaeipour et al. (2008) observed that canola meal has the potential to replace fish meal in diets of rainbow trout without compromising the growth performance. Lim et al., (1997) reported that solvent-extracted canola meal protein are the cost-effective substitutes for fish meal. Buchanan et al. (1997) observed that enzyme treated canola meal in the diets of juvenile *Peneaus monodon*, results in better FCR and higher weight gain compared to the others. Canola meal is well known as a protein ingredient in salmonid diets (Higgs et al., 1996) [21]. According to McCurdy and March, (1992) Solvent-extraction of fiber-reduced canola meal could improve the fish response may be due to the reduced glucosinolate and sinapine level. Soares et al. (2001) suggested 35.40% inclusion of canola meal can replace 48.17% of soybean meal in Nile tilapia diet. According to
Webster et al. (1997) [60], incorporation of canola meal in channel catfish (Ictalurus punctatus) diet is cost effective compare to other plant protein ingredients used in commercial catfish feed.

**Palm kernel meal**

Palm kernel meal is considered as an important alternative feed ingredient in aqua feed production. This oil palm is cultivated mainly for its high oil content. Generally, palm oil is extracted from fruit flesh whereas palm kernel oil is extracted from fruit kernel. Palm oil is known as “poor man’s cooking oil”, commonly used in tropical Africa and in South-East Asian countries (Prabhakaran Nair, 2010) [40]. Palm kernel meal is the by-product formed as a result of the palm kernel oil extraction process. After mechanical extraction, palm kernel meal contains 5-12% oil and kernel meal resulting from solvent-extraction contains 0.5-3% oil (Chin, 2001) [9]. It is fibrous in nature and has medium grade protein, hence suitable for ruminant or rabbit feeding (Pickard, 2005).

Like most of the plant-based ingredients and oilseed meal, there are numerous factors that can limit the inclusion of Palm kernel meal in fish diets. These factors include (1) presence of anti-nutritional factors (2) relatively low protein content (14 to 20% dry weight basis) (Table no 4) and (3) possible amino acid deficiencies. Hence, nutritionally it is inferior to the other oil seed meals such as soybean meal, cottonseed meal and groundnut meal. It is reported that due to the dry and gritty nature, it is not preferred by pigs and ruminants, so it is incorporated in the compound feed (Göhl, 1982) [10]. The incorporation of palm kernel meal, as a fish feed, is restricted due to its less protein content. Ng (2004) reported that 20% dietary inclusion of palm kernel meal in tilapia showed higher growth performance. Omoriege et al. (1993) [47] observed that nile tilapia fingerling fed with 60% palm kernel meal showed similar growth performance as of fish fed with fishmeal-based diet. According to Adikwu (1997) [2], 30 to 90% replacement of fishmeal using palm kernel meal in nile tilapia feed resulted in decreased growth. Ng et al. (2002) [44, 45] reported that commercial feed enzymes treated palm kernel meal incorporated in the diet of red tilapia showed better growth performance than the fish fed with raw palm kernel meal. The study noticed that the inclusion of enzyme-treated palm kernel meal, up to 30%, did not cause any significant decrease in feed utilization and growth of the fish. Ng. (2004) suggested solid state fermentation of palm kernel meal by using fungi like Aspergillus flavus or Trichoderma koningii increase the protein content. In some of the experiments conducted with fermented palm kernel meal in tilapia, protein content and digestibility of palm kernel meal were increased, but a significant reduction in fish growth was noticed, it could be due to the presence of fungal toxins (Ng et al., 2002; Ng, 2004) [44, 45]. Oliveira et al. (1997) reported that pacu fish (Piaractus mesopotamicus) fed with diets containing 70% palm kernel meal showed less digestibility (54%) compared to copra meal. Ng et al., (2002) [44, 45] suggested that, in Hybrid catfish (Clarias macrocephalus x Clarias gariepinus) palm kernel meal @20% could be incorporated without any negative impacts on its growth performance. Hence, palm kernel meal can be used for partial replacement of other high valued protein sources.

**Jatropha kernel meal**

*Jatropha* genus belongs to the family Euphorbiaceae and has approximately 170 known species. *Jatropha curcas* is commonly known as purging nut or physic nut. The average seed weight is 0.64 g and it contains 38–40% of oil indicating its potential as a renewable source of energy (Makkar et al., 1998; Kumar and Sharma, 2008) [33, 56] for bio-diesel production. The *J. curcas* seeds are rich in oil (27.36%) and protein (32.88%), and in addition to that, it also contains a significant amount of macro-minerals (Na, K, Mg, Ca, P) and micro-minerals (Mn, Fe, Zn) (Abou-Arab and Abu-Salem, 2010) [1]. Chemical composition of Jatropha kernel meal are shown in Table no 3. The Jatropha kernel meal, obtained after oil extraction, is an excellent source of nutrients and contains 58–62% crude protein (Makkar et al., 2008). Except lysine, all essential amino acids levels are higher in Jatropha kernel meal than Soybean meal (Kumar et al., 2010) [28]. According to Makkar et al., 2008 [30], high levels of anti-nutritional factors such as trypsin inhibitor, lectin and phytate are present in the seed meal of *J. curcas* and the major toxic component is phorbol esters (PES) (Makkar and Becker, 1997) [35] which restrict their use in fish feed. Anti-nutrients such as protease inhibitors and lectins are heat labile and easy to inactivate by moist heating (Makkar and Becker, 2009) [32]. Areghore et al. (2003) [5] observed that 95% reduction of PEs content in Jatropha kernel meal after heat treatment at 121°C for half an hour and followed by washing with aqueous methanol (92%) (four times). Harter et al. (2011) [120] reported that 50% of fish meal protein in shrimp diets could be replaced by detoxified Jatropha kernel meal, without sacrificing growth and nutrient utilization parameters. Overall, the nutrient utilization and growth performance of white leg shrimp (i.e., L. vannamei) to detoxified Jatropha Kernel Meal fed groups were better than fish meal-fed groups which indicate that shrimp can efficiently use detoxified Jatropha kernel meal. Kumar et al. (2012) [27] concluded that Nile tilapia fingerling fed with control diet containing fishmeal, and the test diets where 62.5% of FM protein was replaced by *Jatropha* and Soybean meal showed no significant difference in feed conversion ratio, protein efficiency ratio and in the growth performance which indicates that *J. phytophylia* kernel meal can be used as a protein source in fish feed. Saha and Ghosh (2013) [49] demonstrated that an inclusion level, up to 30% of fermented defoiled *J. Curcas* seed meal (DJSM), replacing 15% FM could be incorporated in the practical diet for rohu fingerling without compromising growth, feed utilization efficiency and whole body composition. Shamma et al. (2015) [52] compared the nutritional potential of fermented *Jatropha* protein concentrate (FJPC) and soybean protein concentrate. The study found that FJPC detoxified by solid state fermentation could be incorporated up to 20% in the diets of *L. rohita* fingerling without compromising growth and nutrient utilization, but reduction in percent weight gain, protein efficiency ratio and lower survival rate were recorded in non-fermented JPC fed groups. The reason for growth reduction in non-fermented JPC is presence of high level of phorbol ester (1.4 mg g-1). Hence, it can be concluded that solid state fermentation, using *Aspergillus niger*, is an effective method for the removal of PEs in JKM which improves the protein utilization efficiency of JKM.

**Peanut meal**

Solid residues obtained from peanut, after oil extraction is called as peanut meal. It is considered as a rich source of protein containing about 48% protein but deficit in methionine and lysine which limits their incorporation in fish feed. It contains 35 to 40% oil which is extracted by using mechanical and solvent extraction method. It is reported to contain a high amount of arginine (Batal et al., 2005) [4].
Proximate composition of peanut meal is shown in Table 6. Heat treated peanut meal does not contain any anti nutritional factors but they are prone to fungal infestation. Yue et al. (2012) [63] reported that a mixture of Soybean meal and Peanut meal could reduce the inclusion of fish meal in the diet of Litopenaeus vannamei from 300g/kg to 200 g/kg. Liu et al. (2012) [38] suggested that 140g/kg peanut meal can be included in the diet of Litopenaeus vannamei without affecting the growth performance. Inclusion level of peanut meal is reported to be 15 to 20% in catfish diet without supplementation of lysine (Robinson el al., 2001) [49]. In general, the use of peanut meal in the fish diet is not widely practiced due to its amino acid (lysine) deficiency and occasional availability.

Table 1: Protein content and amino acid profile of Soybean meal and soy protein concentrate

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Crude protein (%)</th>
<th>Lysine (%)</th>
<th>Methionine (%)</th>
<th>Threonine (%)</th>
<th>Isoleucine (%)</th>
<th>Tryptophan (%)</th>
<th>Arginine (%)</th>
<th>Phenylalanine (%)</th>
<th>Valine (%)</th>
<th>Histidine (%)</th>
<th>Leucine (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Soybean meal</td>
<td>44</td>
<td>2.83</td>
<td>0.61</td>
<td>1.73</td>
<td>1.99</td>
<td>0.61</td>
<td>3.23</td>
<td>2.18</td>
<td>2.06</td>
<td>1.17</td>
<td>3.42</td>
</tr>
<tr>
<td>Soy protein concentrate</td>
<td>65</td>
<td>4.23</td>
<td>0.91</td>
<td>2.73</td>
<td>3.19</td>
<td>0.78</td>
<td>4.94</td>
<td>3.45</td>
<td>3.38</td>
<td>1.82</td>
<td>5.2</td>
</tr>
</tbody>
</table>

(Adopted from NRC, 2012)

Table 2: Proximate composition of rubber seed meal

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture (%)</td>
<td>16.00</td>
<td>14.30</td>
</tr>
<tr>
<td>Ash (%)</td>
<td>0.24</td>
<td>1.80</td>
</tr>
<tr>
<td>Crude protein (%)</td>
<td>25.40</td>
<td>26.10</td>
</tr>
<tr>
<td>Crude fat (%)</td>
<td>39.10</td>
<td>11.00</td>
</tr>
<tr>
<td>Crude fibre (%)</td>
<td>75.00</td>
<td>43.00</td>
</tr>
<tr>
<td>Nitrogen free extract (%)</td>
<td>33.20</td>
<td>13.80</td>
</tr>
</tbody>
</table>

(Adopted from Bell and keith, 1991)

Table 3: Proximate composition of Canola meal

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Moisture (%)</th>
<th>Ash (%)</th>
<th>Crude protein (%)</th>
<th>Ether extract (%)</th>
<th>Crude fiber (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Iranian canola meal</td>
<td>12.0</td>
<td>6.7</td>
<td>37.57</td>
<td>1.8</td>
<td>13.97</td>
</tr>
<tr>
<td>Canadian canola meal</td>
<td>12.0</td>
<td>6.01</td>
<td>36.0</td>
<td>3.50</td>
<td>11.70</td>
</tr>
</tbody>
</table>

(Adopted from Bell and keith, 1991)

Table 4: proximate composition of palm kernel meal

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Value (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture</td>
<td>8.25</td>
</tr>
<tr>
<td>Crude protein</td>
<td>16.60</td>
</tr>
<tr>
<td>Crude fiber</td>
<td>12.29</td>
</tr>
<tr>
<td>Crude fat</td>
<td>7.59</td>
</tr>
<tr>
<td>Ash</td>
<td>3.88</td>
</tr>
<tr>
<td>Nitrogen free extract</td>
<td>51.39</td>
</tr>
</tbody>
</table>

(Adopted from Ezieszhi et al. 2007)

Table 5: Chemical composition of kernel and shell of J. curcas

<table>
<thead>
<tr>
<th>Constituents(% DM)</th>
<th>Toxic variety</th>
<th>Nontoxic variety</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Kernel Shell</td>
<td>Kernel Shell</td>
</tr>
<tr>
<td>Crude protein</td>
<td>22.2</td>
<td>27.2</td>
</tr>
<tr>
<td>Lipid</td>
<td>57.8</td>
<td>58.4</td>
</tr>
<tr>
<td>Ash</td>
<td>3.6</td>
<td>6.0</td>
</tr>
<tr>
<td>Natural detergent fibre</td>
<td>3.8</td>
<td>3.8</td>
</tr>
<tr>
<td>Acid detergent fibre</td>
<td>3.0</td>
<td>2.4</td>
</tr>
<tr>
<td>Acid detergent Lignin</td>
<td>0.2</td>
<td>0.0</td>
</tr>
<tr>
<td>Gross energy(MJ/kg)</td>
<td>30.7</td>
<td>31.1</td>
</tr>
</tbody>
</table>

(Adopted from: Makkar and Becker, 1999)

Table 6: Proximate composition of peanut meal

<table>
<thead>
<tr>
<th>Peanut meal</th>
<th>Moisture (%)</th>
<th>Crude protein (%)</th>
<th>Crude fiber (%)</th>
<th>Ether extract (%)</th>
<th>Ash (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>After mechanical extraction</td>
<td>7</td>
<td>48.1</td>
<td>6.9</td>
<td>5.8</td>
<td>5.1</td>
</tr>
<tr>
<td>After solvent extraction</td>
<td>8</td>
<td>48.1</td>
<td>9.9</td>
<td>1.3</td>
<td>5.8</td>
</tr>
</tbody>
</table>

(Adopted from National Research Council, 1994)

Conclusion

Has future of fish feed should rely on fish meal as a protein source? The answer is big NO. However, on the other side, demand for fish meal is increasing, among the livestock feed production sector, which hiking the price of fish meal thus ultimately increasing the operational cost of fish farming. So, to meet out the increasing demand of protein rich feed ingredient, aqua feed production sector must find an alternative options to fish meal. Plant based protein sources could be the possible solution, since, it is easily available and rich in protein. However, it is not a complete solution to that question, since, it contains heat-labile and heat-stable anti nutritional factors thus limiting their incorporation in fish feed by affecting the growth performance and metabolic activity of the fish. These anti nutritional factors can be reduced or eliminated by different simple detoxification methods such as heating, solvent extraction, chemical treatment, fermentation, etc., in order to enhance the digestibility and nutritional quality of the ingredient. In future, in-situ based techniques – plant protein utilization efficiency should be increased by incorporating enzymes/bacteria which can stabilize the anti-nutritional factors of plant in fish gut itself – need to be developed to improve the plant based protein feed ingredient in fish feed.

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