

# Consequences of Replacing Fish Oil with Vegetable Oils in Fish

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## Abstract

Due to the continuous demand for aquatic products such as fish and shellfish coupled with the continuous expansion of aquaculture there is the need for better and sustainable feed. However, fish oil which is used traditionally as the main lipid source has seen a reduction in its production hence stakeholders seeking for a new suitable lipid source to be used in place of the traditional fish oil. In this study we review the impact vegetable oils have on growth and feed utilization of economically important fish species. Also we report on how vegetable oils when used in place of fish oil affect immune system and health of fish as well as the proximate composition and fatty acids. Finally we look at how it affects lipid metabolism related genes in fish. This paper documents that using vegetable in place of fish oil has the tendency of increasing aquaculture production as well as maximizing profit. It is clear from studies conducted so far that different vegetables such as palm oil, coconut oil, sunflower oil and a host of other plant based oils can be used in place of fish oil without any compromise on growth even though it modifies the fatty acid and body composition to some extent.

Owing to over-exploitation of fish for its oil and meal [5], there are calls for alternatives [6]. Plant oils have been considered to be suitable alternative to replace FO. This is because they are cheaper and have higher resistance to lipid peroxidation [7]. They however, lack long chain polyunsaturated fatty acids (LC-PUFA), such as eicosapentaenoic (EPA, 20:5n-3), docosahexaenoic (DHA, 22:6n-3), and arachidonic acids (20:4 n-6) (ARA) which are required in the fillets for a healthier product for human consumers [8]. Several studies have been conducted in attempt to replace fish oil with suitable alternatives [9-11]. Turchini, et al. [12] reviewed replacement of fish oil in fin fish. Also, [13] reviewed the benefits of fish oil replacement by plant originated oils in compounded fish feeds. These studies however, did not emphasize on how replacing fish oil with other oil sources influence lipid metabolism related genes as well as immune system of the fish. The aim of this paper is therefore to present updated review focusing on how vegetable oils affects genes involved in lipid metabolism. This review gives a current update on the effects of vegetable oils on growth of fish, feed utilization, body composition and fatty acid composition as and further discuss how replacing fish oil with vegetable oils influence lipid metabolism related gene expression and immune system of fish.

**Keywords:** Fish oil; Vegetable oil; Growth regulation; Lipid metabolism; Genes; Review

## Introduction

As a result of the rapid development of intensive aquaculture, it is essential to produce good quality feeds that can last over a longer period [1]. In aqua feeds, lipids are important since they aid in the transportation of fat-soluble vitamins as well as their absorption. Because fish oil (FO) provides essential polyunsaturated fatty acid (PUFA), especially high unsaturated fatty acid (HUFA) [2], they have been the main source of lipid in aqua feeds [3]. However, FO cannot meet the increasing demand of aqua feed industry due to the decline in its production [4].

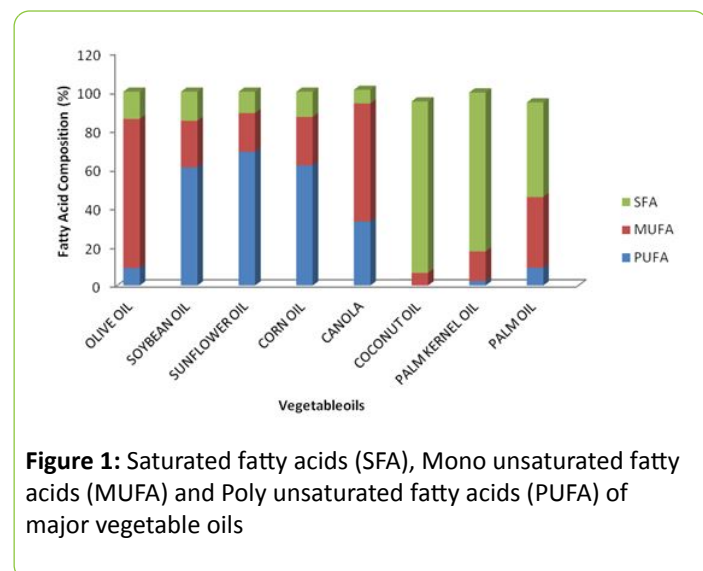
## Methodology

### Article collection (selection)

To evaluate the consequence of substituting fish oil with vegetable oils on various fish species, English databases such as Springer, Wiley, Elsevier etc were browsed from 1 January 2007 to 10 August 2017. For the purpose of this study, the present search was done using aquaculture nutrition subject headings, terms and a combination of several keywords including: "Replacement"; "Vegetable oil"; "Palm oil"; "Coconut oil"; "Tilapia"; "Fish oil"; "Canola oil"; "Cat fish"; "Sea bream"; and "Rainbow trout".

## Vegetable oils used in fish diets

**Qualities of vegetable oils used in fish feed:** Vegetable oils have been used extensively in aquaculture feeds. They include; palm oil, coconut oil, soybean oil, corn oil, camelina oil, perilla oil, cotton seed oil and a host of others. In this section we review the characteristics of vegetable oils that are of great potential in aquaculture feed production. Although, vegetable oils have different fatty acids, they generally lack n-3 LC-PUFA but have higher amounts of other fatty acids such as n-3 PUFA, n-6 PUFA, MUFA and SFA. It is reported that olive oil, rapeseed and canola oil are rich in MUFA while sunflower oil, corn oil and soybean oil possess higher quantities of n-6 PUFA. Also, oils such as palm oil and coconut oil are rich in SFA with linseed oil and perilla oil being abundant in n-3 PUFA [12,14]. The fatty acid composition (SFA, MUFA and PUFA) of some selected vegetables have been shown in **Figure 1**.



Also, **Table 1** gives a summary of other fatty acid (C: 16, C: 18, C 18:1, C18:2 and C18:3) composition of some selected vegetable oil as reported by oil palm knowledge base in 2014. Palm oil, Sunflower oil, Olive oil, Safflower oil and Linseed oil have higher levels of C:16, C:18, C18:1, C18:2 and C18:3 respectively in the values of 44, 5, 78, 75 and 60 respectively.

**Palm oil:** Palm oil is unique among vegetable oils because of its high saturated acid content with a significant amount (10%-16%) of saturated acids at the 2-position of its triglycerides. Palm oil also contains appreciable amounts of diglycerides (5%-8%) and free fatty acids, which can have a substantial effect on its physical properties. PO contains 50% SFAs, mostly palmitic acid (PA, 44%) and lower amounts of stearic acid (5%), 40% monounsaturated fatty acids (MUFAs), mostly oleic acid, and 10% polyunsaturated fatty acids (PUFAs), mostly linoleic acids [15-17].

**Coconut oil:** Coconut oil (CO) is made up of approximately 40%-45% MCFA with lauric acid (C12:0) of total fatty acids. Also, CO has a higher amount of saturated fat content and has a high melting point. This characteristic makes it stable to heat and can withstand peroxidation, allowing its long-term storage [18]. Coconut oil is regarded as cheaper, more sustainable, readily available and one of the most stable oils with high resistance to oxidative rancidity and ability to withstand warm temperatures unlike other oils [19].

**Soybean oil:** At present, soybean oil (SO) is widely used in freshwater aqua-feeds because its production is steadily increasing and has reasonable prices. SO is rich in PUFAs, especially LA which is EFAs for freshwater fish [20,21]. Soybean oils contain 24% MUFA (Oleic), 61% PUFA (Linoleic) as well as 15% SFA. Also, it has an SFA:MUFA:PUFA ratio of 15:24:58.

**Table 1:** Fatty acid (C:16, C:18, C18:1, C18:2 and C18:3) composition of some selected vegetable oils.

Vegetable oil	C16:0	C18:0	C18:1	C18:2	C18:3
Corn	13	3	31	52	1
Cotton seed	23	2	17	56	*
Linseed	6	3	17	14	60
Olive	10	2	78	7	1
Pam oil	44	4	39	11	*
Safflower	7	3	14	75	*
Soybean	11	4	23	53	8
Sunflower	6	5	20	60	*

\* Values less than 1. Source: Oil Palm Knowledge Base, 2014

**Camelina oil:** *Camelina sativa*, belongs to the Brassicaceae family. It has higher content of ALA. It is estimated to contain about 45% of ALA [22]. It serves as a good substrate for the biosynthesis of n-3 LC-PUFA.

**Perilla seed oil:** Perilla oil has higher content of n-3 fatty acids, approximated to be between 55% and 66% LNA [23]. Also, it serves as a precursor for the biosynthesis of long-chain fatty acids of the omega-3 series which includes DHA and EPA [24].

**Linseed oil:** Linseed oil, a precursor of the LC n-3 PUFA series is ranked as one of the world's vegetable oils rich in LNA [25]. It contains very high proportions of linolenic acid (18:3n-3).

**Echium oil (EO):** Relatively, Echium seed oils has LNA and 18:4n-3 (stearidonic acid, SDA) in higher quantities. It also has a higher proportion of 18:3n-6 ( $\gamma$ -linolenic acid, GLA) [26]. Comparing to other vegetable oils, EO has moderate levels of LA [27,28] and a well-balanced n-3/n-6 PUFA ratio.

**Canola oil:** Canola has a higher proportion of ALA [29]. Canola oil is known to possess an n-3/n-6 ratio of about 1:2 [30]. Canola oil are abundant in fatty acids with 18 carbons, specifically monounsaturated fatty acids, ALA, LA and OA. It is however, low with respect to saturated FA. Generally it has low amount of erucic acid [31].

**Peanut oil:** Peanut oil is rich in monounsaturated fatty acids [32]. Approximately 51%, 30% and 19% of the oil are monounsaturated, polyunsaturated and saturated fatty acids respectively. Palmitic oleic and inoleic acids makeup almost 40-50% of the entire fatty acids. In addition, arachidonic acids, behenic and other important fatty acids make up 6-8% of the fatty acids.

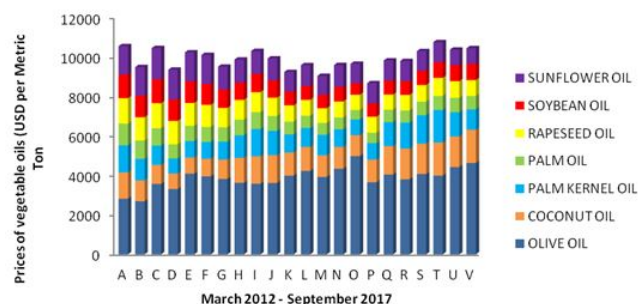
**Palm kernel oil:** Palm kernel oil contains about 81% saturated fats [33]. It has high levels of lauric acid (C12:0) and has slightly less of the short-chain fatty acids and more oleic acid and saturated fatty acids that are short and medium chain fatty acids such as caprylic and capric acid.

**Cotton seed oil:** Cottonseed oil (CSO) is ranked fifth in terms of total vegetable oils produced globally. As shown in **Table 1**, cotton seed oil is rich in C18:2 fatty acid (52%) as well as C18:1 fatty acid (31%). It however has low quantity of C18:3 fatty acid (1%). Also, it contains high concentration of linoleic acid (LA, 18:2n-6) [34].

**Prices of vegetable oils:** The prime aim of extensive as well as semi-intensive aquaculture is to maximize profit. One major factor that has the tendency to affect profit in the aquaculture is the choice of oil used in fish feed. The different oils have different prices and choosing one that has lower cost and affects growth positively is considered suitable for fish feed. Comparatively vegetable oils are known to be cheaper. Below are the prices of some vegetable oils used in fish feed as reported by Mundus Index (**Figure 2**) [35].

Amongst the 7 types of vegetable oils in **Figure 2**, olive oil is the most expensive. The highest price recorded amongst the vegetable oils was recorded in September, 2015 with an amount of 4,986.87 MT. The least price was however recorded by palm oil in September, 2015 with an amount of 483.49 MT. Generally, the prices of vegetable oils such as coconut oil, palm kernel oil, palm oil, soybean oil and sunflower oil do not differ much and are cheap.

**Production of vegetable oils:** Vegetable oils have been touted as suitable alternatives to fish oil in aquaculture feeds, not only because their relatively cheaper prices but also due to their stable production. The production volume of the major vegetable oils is shown in **Figure 3**.

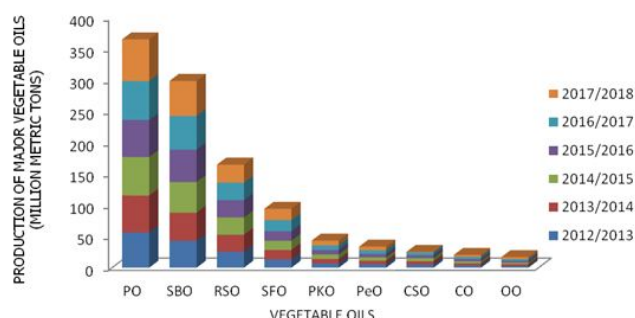


**Figure 2:** Quarterly prices of vegetable oils from March 2012-December 2016. A-Mar 12; B-Jun 12; C-Sep 12; D-Dec 12; E-Mar 13; F-Jun 13; G-Sep 13; H-Dec 13; I-Mar 14; J-Jun 14; K-Sep 14; L-Dec 14; M-Mar 15; N-Jun 15; O-Sep 15; P-Dec 15; Q-Mar 16; R-Jun 16; S-Sep 16; T-Dec 16; U-Mar 17; V-Jun 17 (source: Mundus Index, 2017 [35]).

The production of vegetable oils has over the years continued to be stable with increase in volume. In 2016/2017, 66.32 and 53.94 metric tons of Palm oil (PO) and Soybean oil (SBO) respectively were produced. It has however, been projected that 66.86 and 56.13 million metric tons of PO and SBO respectively will be produced in 2017/2018 production year. Although CSO declined from 4.48 to 4.41 million metric tons in 2016/2017, it is expected to increase to 4.99 million metric tons will be produced in the year 2017/2018. Also, PKO is expected to rise from 7.33 million metric ton in 2016/2017 to 7.81 million metric tons in 2017/2018. Of the seven major vegetable oils, the least produced vegetable oil is olive oil.

**Effects of vegetable oil on growth, feed utilization and survival of fish:** Attempts have been made to use vegetable oils in place of fish oil over the past years by fish nutritionists with emphasis on how these lipid sources affect the growth performance of fish as well as their ability to utilize the various diets. Replacing FO with VO are generally known not to have negative effects on several fish species such as Black carp [36], Atlantic salmon [37], Hybrid sturgeon [38], Cobia [39] and Murray cod [40]. There are instances where substituting FO with VO either complete or partial improved growth performance, feed utilization and survival of some fish species such as *Siganus canaliculatus* [10] and *Pangasius hypophthalmus* [41]. This is probably because the VOs were able to provide the required amount of fatty acids that are essential for growth.

Peng et al. [42] replaced fish oil with soybean oil in juvenile turbot and reported that, growth performance was not compromised irrespective of the oil source. They reported fish oil could be replaced with soybean oil at 66.7% to improve growth. Similarly, Sun et al. 2011 [36] in a feeding trial used soybean oil in place of fish oil. They fed rabbit fish with four diets with 0%, 23%, 45% and 67% soybean. The best growth performance was obtained when fish oil was replaced with 45% soybean oil.



**Figure 3:** Production of major vegetable oils worldwide from 2012/2013 to 2017/2018 by type. (Million metric tons). PO: Palm oil; SBO: Soybean oil; RSO: Rapeseed oil; SFO: Sunflower oil; PKO: Palm kernel oil; PeO: Peanut oil; CSO: Cotton seed oil; CO: Coconut oil; OO: Olive oil.

**Table 2** gives a summary of studies on how substituting fish oil with different vegetable oils affects growth and feed utilization of commercially important fish species.

These studies were in agreement to previous study by Guller and Yildiz, [34] that recorded optimal growth and feed utilization when 50% cottonseed oil was used as a substitute for fish oil in rainbow trout.

Similarly, Demir et al. [43] evaluated the effects of partial or total replacement of fish oil (FO) with unrefined peanut oil (PO) on growth and some physiological parameters using juvenile *Mozambique tilapia* (initial weight  $6.36 \pm 0.19$  g) as the experimental fish for 60 days. Group fed diets containing 50% PO recorded higher values of weight gain as well as feed efficiency compared to other groups.

A 60-day growth experiment was carried out by Singh, et al. [44] to evaluate the impact of replacing dietary cod liver oil

(CLO) with palm oil (PO) on the growth and survival of the Indian major carp, *Cirrhinus mrigala*. In this study, Singh et al. [44] fed *C. mrigala* fry ( $2.16 \pm 0.01$  g) with five experimental diets with composition as follows; 100% CLO+0% PO (control), 25% PO +75% CLO, 50% PO+50% CLO, 75% PO+25% CLO, and 100% PO +0% CLO. The results showed that growth performance and survival were not altered when fish oil was substituted with PO up to 25% but growth performance was retarded when PO was increased.

Sonu et al. [45] conducted a study to assess the impact of using sunflower oil (SFO) instead of fish oil has on survival and growth parameters of *Cyprinus carpio* (Linn.) using five isonitrogenous and isoenergetic dietary treatments (0%, 25%, 50%, 75% and 100% replacement of fish oil with sunflower oil) over a period of 60 days. Substituting fish oil with sunflower oil did not affect both weight gain and specific growth rate significantly. Also, survival was indifferent with respect to using sunflower oil in place of fish oil.

From literature, results of previous studies on different species [8,12,36,46-50] documented that using various vegetable oils to substitute fish oil either in whole or in part does not compromise growth as well as feed utilization.

These studies mentioned above as well as in **Table 2**, points out that fish oil can be replaced with vegetable oils partially or wholly either by use of single vegetable oils or as blend. It is however worth noting that the effects these vegetable oils have on growth performance and feed utilization is subject to factors such as age and size of fish and the species under consideration. Also the characteristics of the vegetable oil determine the extent to which it could be used in place of fish oil. Also, the particular essential fatty acid requirements of the studied species, the dietary inclusion of fish meal or other fatty acid sources and the lipid content of diets assayed for each species and the ability to accept vegetable oils of the target fish could be the underlining factors resulting in the above differences.

**Table 2:** Effects of vegetable oils on growth and feed utilization in fish.

Vegetable oil	% of fish oil substitution	Species	Initial weight (g)	Effects on growth and feed utilization	References
Palm oil	0, 33.33, 66.67, 100%	<i>Oreochromis niloticus</i>	9.09	Palm oil can effectively replace cod liver oil in diets for <i>Nile Tilapia</i> , <i>O. niloticus</i>	[9]
	0, 25, 50, 75, 100%	<i>Cirrhinus mrigala</i>	2.16	Palm oil can substitute fish oil up to 25% without compromising growth	[44]
	N/A	<i>Heterobranchius longifilis</i>	5.53	Palm oil can replace fish oil in diet of catfish totally without any adverse effects.	[51]
	N/A	<i>Larmichthys crocea</i>	245.29	Replacing fish oil with palm oil up to 100% does not compromise growth and feed efficiency.	[52]
		<i>Clarias gariepinus</i>	15.14	Palm oil is a good substitute in diets of catfish.	[53]
Soybean oil	0, 60, 80, 100%	<i>Acanthopagrus schlegelii</i>	20.26	Placed with soybean oil up to 80%. A complete replacement results in reduced growth performance	[54]
	0, 25, 75, 100%	<i>Pelteobagrus vachelli</i>	1	Soybean oil can be used in place of fish oil up to 25%	[55]

	0, 20, 32, 56, 100%	<i>Megalobrama amblycephala</i>	0.3	Replacing fish oil with 20% soybean oil in Blunt snout bream juvenile diet enhances growth and feed utilization	[21]
	33.3, 66.7, 100%	<i>Scophthalmus maximus L.</i>	5.88	Soybean oil can be used in place of fish oil up to 66.7% for maximum growth and feed utilization	[42]
	100%	<i>Larmichthys crocea</i>	13.77	Replacing Fish oil with soybean oil at 100% can increase growth and feed efficiency	[56]
Flaxseed oil	0, 25, 50 and 75%	<i>Anoplopoma fimbria</i>	153.4	Replacing fish oil with flaxseed oil up to 75% enhanced growth as well as feed utilization	[57]
	0, 50 and 100%	<i>Onchorhynchus mykiss</i>	16.5	Feeding <i>O. mykiss</i> with 50% and 100% flaxseed oil doesn't retard growth	[58]
Cotton seed oil	0, 50, 60 and 70%	<i>Sparus aurata</i>	4	Fish oil replacement by Cotton seed oil up to 60% level had no detrimental effects on growth or nutritive utilization	[50]
Peanut oil	0, 50, and 100%	<i>Oreochromis mossambicu</i>	6.36	Peanut oil can replace fish oil up to 50% without affecting growth performance adversely	[43]
	0, 25, 50, 75 and 100%	<i>Oncorhynchus mykiss</i>	N/A	Peanut oil can replace fish oil up to 50% without affecting growth performance adversely	[59]
	100%	<i>Acipenser gueldenstaedtii</i>	1.39	Total replacement of fish oil with fish oil enhanced growth and feed utilization	[1]
Linseed oil	0, 25, 50, 75 and 100%	<i>Oreochromis niloticus</i>	22	Linseed oil can replace fish oil up to 50% for maximum growth. A further increase declines growth	[1]
	33.3, 50, 66.7	<i>Puntius gonionotus</i>	12.1	Replacing fish oil with linseed oil up to 33.3% does not compromise growth	[60]
Echium oil	0 and 50%	<i>Sparus aurata L.</i>	265.05	Feeding Gilthead sea bream with 50% Echium oil does not compromise growth and feed efficiency	[61]
Rapeseed oil	0 and 100%	<i>Dicentrarchus labrax</i>	35.4	Total replacement of FO with RO improves growth and feed utilization	[62]

### Effects of vegetable oils on proximate and fatty acid composition:

Several studies have been conducted with respect to how alternative plant oils affect the body composition as well as fatty acid profiles. Replacing fish oil with vegetable oils have been reported to alter the fatty acid profile of the end product and to some extent influence the quality of the fish significantly [13,14,63]. In a study by Nasopoulou et al. [13], sing canola oil instead of fish oil in diets of Jade Tiger hybrid abalone affected muscle fatty acid composition. From the results, docosahexaenoic acid (DHA, 22:6n-3), omega-3 polyunsaturated fatty acid (n-3 PUFA) as well as eicosapentaenoic acid (EPA, 20:5n-3) were elevated in abalone groups fed diets containing 25% and 50% canola oil compared to the group fed canola oil free diets. The highest levels of total PUFA and total monounsaturated fatty acids occurred in the 100% CO group. Similarly, replacing 8% fish oil olive pomace resulted in a reduction in the EPA and DHA content in gilthead sea bream. It is interesting to note that in most cases, the fatty acid composition of muscle, liver as well as whole body reflect the dietary fatty acids [7,62-64]. Despite the general phenomenon of tissue fatty acid corresponding with their dietary fatty acids, there are instances where diets did not necessarily affect the tissue fatty acids. Factors which includes but not limited to nutritional status of the fish at stocking, differences in species and size as well as

feeding duration could be the reason for difference in results of the studies [55].

With respect to proximate composition, ash, protein, lipids as well as carbohydrates have been the major parameters. There have been different results with some scientist reporting that replacing fish oil with vegetable oils affects proximate composition whiles others have reported otherwise. When rapeseed oil was used in place of fish oil in diets of Atlantic salmon, there was an increase in whole body protein as well as protein retention [65]. This could be attributed to the presence of mid-chain fatty acids such as OA in vegetable oils which are oxidized efficiently and used by fish for the production of adenosine tri-phosphate for energy purposes [2,12]. On the contrary, there are studies reporting replacing fish oil with vegetable oils does not influence body composition. For instance, Li et al. [66] documented that linseed oil as a replacement for *tilapia* did not vary whole body protein, lipid and ash contents and is similar to the study in silver barb where replacement of FO with LO yielded n changes in proximate composition [60]. In other studies, inclusion of vegetable oil (soybean oil) as FO substitute in the diets of black sea bream [54], gilthead sea bream [67] and *turbot* [42] also did not influence the whole body proximate compositions.

These contradictions could be attributed to the length of study, lipid sources, lipid levels, type and size of fish as well as culturing conditions. The type of feed (pelleted or extruded) is known to influence the proximate composition [68].

**Effects of vegetable oils on serum metabolites, antioxidant capacity, immune response and health of fish:** In order to ensure a sustainable aquaculture in the long term, there is the need for fish with good immune function and health [69]. There is therefore the need to ensure nutrition of fish is geared towards the production of feed that can boost the immune system of fish. Using alternative lipid sources should not only aim at supplying lipids at the appropriate levels that will balance EFAs for growth but also for proper immune function. Due to this idea, there have been some studies over the years on how the use of VOs could influence the health and immune function of fish using a wide range of parameters.

Blood parameters, serum composition and biochemical constituents are the most indicator of the general condition of the fish [70]. The general status of fish health is mostly determined by the use of serum biochemical parameters. Blood analysis is a useful tool that can provide reliable information on metabolic disorders, deficiencies, adaptation processes to various environmental influences and chronic stress status [71]. Fish hematology parameters have gained importance in aquaculture simply because it is used in monitoring the health status of fish. Ochang [11] reported that hematological parameters were significantly influenced when soybean oil was used as a substitute for cod liver oil in Nile tilapia, *Oreochromis niloticus*. Red blood cell, and Total cholesterol were altered significantly while total protein, glucose concentration, AST and ALT were significantly altered when palm oil was used as a substitute for fish oil in African catfish fingerlings [51]. The above results are indicative that feeding different species of fish with different vegetable oils could influence hematological values. Scientific evidences gathered over the past thirty years indicate that dietary nutrients as well as additives could stimulate the immune system of fish and help to fend off diseases [72]. In fish, the non-specific immune system is more important for disease resistance than specific immune system [73]. Substituting FO with VO affects several functions such as the production of eicosanoid [74] and normal function of immune cells [69]. It is also known to alter the fatty acid composition of cells responsible for immune function [75] as well as humoral immunological processes of which include serum lysozyme activity or alternative complement activity [76]. In addition to the above, the expressions of immune related genes are altered when VOs are used in place of FO [77,78]. These changes might be due to an imbalance of n-3/n-6 ratio when FO is replaced by VO.

The effects of replacing fish oil with unrefined peanut oil on serum indicators were studied in common carp (*Cyprinus carpio*). Carp with initial weight of 29.57 were fed diets with 34.5% Protein and 12.8% lipid for 60. The diet includes semi and total FO replacement by PO. The results of this study showed that serum total protein and globulin level increased with increasing PO in the diets. Triglyceride levels significantly increased in fish feed included 100% PO [49]. This

study is of importance to the use of vegetable oils (peanut oil) in carp aquaculture since there was no compromise in serum total protein and globulin which are used as indicators of how strong or weak the innate immune response of fish are [79]. The interesting aspect is that other studies had also reported no adverse effects of vegetable oils in other fish species such as largemouth bass [80] and caspian brown trout [81]. In a similar study to assess the impact of substituting dietary fish oil with unrefined peanut oil in tilapia, the results indicated PO has great prospects in boosting the immune functions of tilapia. Nutritional status is known to influence the glucose metabolism pathway. Demir et al. [43] documented a decrease in serum glucose when FO was substituted with 50% and 100% PO in tilapia diets. Also, serum cholesterol which is used to determine the health status of fish was lower in the groups fed 50% and 100% PO. In order to ensure cultured fish species are healthy, there is the need to maintain a pro-oxidant-antioxidant balance [82]. In times of pathogen invasion, the biological system of fish like all other organisms need to be protected against ROS-mediated damage. Enzymes such as glutathione S-transferase (GST), glutathione peroxidase (GPx), catalase (CAT) as well as superoxide dismutase (SOD) are responsible for the protection of the fish against ROS-mediated damage [83]. In a study by Wang et al. [84], replacing dietary FO with graded levels of linseed oil in diets of juvenile turbot (*Scophthalmus maximus* L.) did not significantly influence serum total SOD activity. A similar study by Lin and Shiau [85] reported higher levels of serum SOD and CAT in groupers fed a blend of FO and corn oil. These results were in agreement with a study by Sun et al. when dietary fish oil was replaced with rapeseed oil in diets for black carp fingerlings.

In other study, substituting fish oil with linseed oil or soybean oil did not affect serum lysozyme activity in gilthead sea bream (*Sparus aurata*) [86]. Similarly, Geay, et al. [87] documented a non-significant difference in lysozyme activity in f Eurasian perch when linseed oil was used to replace fish oil. Again the report of Mozanzadeh et al. [88] was in agreement with the above studies when Sparidentex hasta juveniles fed diets with either canola oil or sunflower oil in place of fish oil did not affect serum lysozyme activity even though blending fish oil with the two vegetable oils significantly increased lysozyme activity. The higher lysozyme activity in diets with both fish oil and vegetable oils could be as a result of the optimal n-3 and n-6 LC-PUFA ratio. Other studies such as Mourente et al.; Yildirim-Aksoy et al.; Montero et al., [89,90,69] have reported that alternative lipid sources did not alter serum lysozyme activity.

Feeding tilapia with vegetable oils increased the production of SOD. This could be inferred from the study of Li et al. [1] when serum SOD increased with the inclusion of linseed oil. Despite the positive results (non-significant differences or increase in serum SOD, LYS, CAT etc) as reported above, other studies have reported instances where dietary vegetable oils significantly influenced these parameters negatively. Lin and Shiau [85] reported that the plasma lysozyme activity was significantly increased when fish oil was partly (25% and 50%) replaced by corn oil in the diets of grouper (*Epinephelus molabarius*).

**Effects of vegetable oils on lipid metabolism related gene expression of fish:** Different results have been reported as to the effects vegetable oils have on gene expression in different species of fish. Nayak, et al. [60] documented higher mRNA expression levels of  $\Delta 6$  fad in liver, muscle and intestines of silver barb fed on LO diets as compared to other group fed FO diets. This result was not different from a previous study conducted by Ren et al. [91] in common carp. In that study, the levels of  $\Delta 6$  fad in liver and intestine were induced when fish were subjected to LO while the fish fed with FO, had a suppressed expression of  $\Delta 6$  fad. In a recent study Ayisi & Zhao [63] reported that replacing fish oil with palm oil in diets of juvenile tilapia (*Oreochromis niloticus*) affected some genes that responsible for lipid metabolism. By this they reported a positive correlation between dietary PO and with FAS, ACC, SCD1 and ACYL mRNA expression. Also, Li et al. [66] substituted fish oil with soybean oil and documented that increasing dietary soybean oil level up-regulated acyl-CoA delta-9 desaturase and down-regulated peroxisome proliferator-activated receptors- $\alpha$  and  $-\beta$  in blunt snout bream (*Megalobrama amblycephala*) juvenile. It has been documented that PPAR- $\alpha$  and PPAR- $\beta$  are important during the metabolism of lipids [92]. Through the regulation of regulation of the expression of several enzymes involved in the oxidation of fatty acids, PPAR- $\alpha$  catabolise fatty acids [93]. The study of Li et al. again demonstrated that the highest expressions of PPAR- $\alpha$  and PPAR- $\beta$  were detected in the liver of fish fed the 20%SO diet and was down-regulated in those fed dietary SO levels from 20% to 100%.

Substituting FO with VO led to the down regulation of LXR expression in gilthead sea bream (*Sparus aurata* L.) [94] confirming a previous study by [95] which also reported the down regulation of LXR mRNA expression of the liver of salmonids fed VO. LXR plays essential roles in the metabolism of fatty acid, cholesterol and glucose metabolism [96] as well as serving as a cholesterol sensor [96]. Fatty acids influence the regulation of LXR [97]. The effects of substituting fish oil with vegetable oils such as palm oil (PO), rapeseed oil (RO), soybean oil (SO) or linseed oil (LO) on the mRNA expression of genes involved in lipid metabolism were evaluated by Peng et al. [98]. In this study, juvenile turbot (*Scophthalmus maximus* L.) (mean initial body weight,  $9.49 \pm 0.03$  g) fed the different diets with required n3 LC-PUFA for 12-weeks. Compared to fish oil (FO), fish fed SO had significantly higher expression of PPAR $\gamma$  while those fed PO had higher expression of PPAR $\alpha$  and FAS. In addition, they reported an upregulation of ApoB-100, LXR and LPL in fish fed RO and LO compared to those fed FO. They concluded that based on their results, fish ingesting or consuming diets with larger amounts of MUFA as in the case of RO could be a recipe for liver lipid deposition which might have been as a result of the up-regulation of genes involved in the synthesis of fatty acids. These studies as presented above are important to the aquaculture industry since an understanding of how these genes involved in lipid metabolism is important to ensure sustainable aquaculture. Lipid metabolism is complex and knowing how these genes regulate fatty acid utilization and growth is necessary.

It is worth noting that the expression patterns of the above studies differ since some of the genes are up-regulated while

others are down-regulated. These differences observed in results with respect to expression profiles might have arisen due to the differences in species, length of feeding trial as well as nutritional status of diet.

## Conclusion and Future Perspective

From above literature, it could be concluded that fish oil can be replaced with vegetable oils to a large extent. In most cases, replacing fish oils with different vegetable oils single handedly or as a blend did not compromise growth. However these were subject to the species and age of fish under consideration, the duration of the study as well as the amount of n-3 LC-PUFA available in the diet. In cases where there are sufficient amount of n-3 LC-PUFA, there were no compromise on growth. It could also be concluded that the fatty acid composition of fish muscle, liver as well as whole body usually reflects that of the dietary fatty acids, hence the fatty acids of these tissues are easily altered when vegetable oils are used in place of fish oil.

Even though much work has been done with respect to using vegetable oil in place of fish oil, there seem to be limited information with respect to the effects vegetable oils have on the genetic make-up of the various fish species as well as the health status of the various fish species. This could be attributed to the large number of vegetable oils that are of great potential as well as the numerous fish species that are being researched. In recent times, new advancements in nutrition research have allowed for the combination of nutrition and genomics analysis through the nutrigenomics approach, which has added to the understanding of the effect of diet on gene expression [99]. Nutrigenomics – the discipline overarching the regulation of genetic signals by nutrients – holds potential for nutritional innovations and for optimizing animal health, quality and performance, reducing feed costs and, ultimately, fostering sustainability of aquaculture [100]. This trend of nutritional studies allows details studies on how different nutritional components influence the genetic make-up of fish. Studies should therefore be geared towards this angle to understand the molecular basis for growth, immune response as well lipid or fatty acid utilization. In addition to directing nutritional studies towards nutrigenomics, studies should not be focused on a particular life stage (age group). The literature reviewed above shows that most studies were conducted using juveniles. However, it has been shown that the efficiency at which nutrients are utilized, gene expression patterns as well as body composition change along the life span of fish [101]. Studies on replacement of fish oil with vegetable oils should therefore cut across all life stages.

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