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REVIEW: THE VARIOUS SOURCES OF FATTY ACIDSAS A FISH FEED TO INCREASE OMEGA-3 IN CATFISH (*CLARIAS*SP)

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ABSTRACT

The unsaturated fatty acids like omega-3 fatty acids such as linolenic acid, EPA and DHA play an important role for human health. Omega-3 fatty acids can prevent various cardiovascular diseases and improve brain development in children. Omega-3 fatty acids can be synthesized in the human body and animals including fish. The body can synthesize omega-3 fatty acids if it consumes foods that contain high linolenic acid and linoleic acid. One source of animal fatty acids is from fish. Catfish is a low economic fishery commodity and has good prospects for development. Catfish can be cultivated in a limited area with high stocking density. Catfish have higher omega 6 content compared to omega-3 fatty acid content. The Omega-3of catfish is affected by the age and weight of the fish. Catfish are a group of freshwater fish that have higher omega-3 content compared to other freshwater fish. The content of omega-3 fatty acids in catfish can be increased by modifying the formulation in fish feed. Fish feed can be formulated with various raw materials that contain high omega-3 fatty acids. Sources of raw materials for omega-3 fatty acids that can be used as additional ingredients in fish feed are macroalgae, microalgae and soybeans. The raw material can be formulated in fish food in the hope that it can produce fish that have high omega-3 fatty acid content. Catfish are a group of freshwater fish that have higher omega-3 content compared to other freshwater fish. The content of omega-3 fatty acids in catfish can be increased by modifying the formulation in fish feed. Fish feed can be formulated with various raw materials that contain high omega-3 fatty acids. Sources of raw materials for omega-3 fatty acids that can be used as additional ingredients in fish feed are macroalgae, microalgae and soybeans. The raw material can be formulated in fish food in the hope that it can produce fish that have high omega-3 fatty acid content. Catfish are a group of freshwater fish that have higher omega-3 content compared to other freshwater fish. The content of omega-3 fatty acids in catfish can be increased by modifying the formulation in fish feed. Fish feed can be formulated with various raw materials that contain high omega-3 fatty acids. Sources of raw materials for omega-3 fatty acids that can be used as additional ingredients in fish feed are macroalgae, microalgae and soybeans. The raw material can be formulated in fish food in the hope that it can produce fish that have high omega-3 fatty acid content. Fish feed can be formulated with various raw materials that contain high omega-3 fatty acids. Sources of raw materials for omega-3 fatty acids that can be used as additional ingredients in fish feed are macroalgae, microalgae and soybeans. The raw material can be formulated in fish food in the hope that it can produce fish that have high omega-3 fatty acid content. Fish feed can be formulated with various raw materials that contain high omega-3 fatty acids. Sources of raw materials for omega-3 fatty acids that can be used as additional ingredients in fish feed are macroalgae, microalgae and soybeans. The raw material can be formulated in fish food in the hope that it can produce fish that have high omega-3 fatty acid content.

Keyword: Catfish, omega-3, microalgae, macroalgae, soybean oil

Introduction

At present, a pandemickCovid-19 is sweeping the world, with 180 countries with confirmed virus infection of more than 2 million people as of May 2020, and then to resist infection from this virus the immune system of the human body must be increased. Therefore, it is sought a way so that immunity is strong built up, including nutritional intake must contain omega-3 fatty acids, vitamins and probiotics. Food ingredients that contain the highest omega-3 fatty acids are fish, then oysters, shrimp, lobsters, the next order is beef, chicken and mutton. Sea fish, for example mackerel (Rastrelligersp), costs Rp. 45 000 / kg, containing high omega-3 fatty acids. Freshwater fish, for example catfish (Clarias, sp), are more affordable at Rp 35 000 / kg, containing more omega-6 fatty acids, so we will look for ways to make catfish with high levels of omega-3 fatty acids.

Various percentage ofSoybean oil in catfish feed as an additive resulted that soybean oil concentration of 10% in feed will produce catfish oil with the highest omega-3 fatty acids, EPA and DHA [1].Oils from catfish flour have a fairly good fatty acid profile because it contains oleic fatty acid (C18: 1) of 22.65%, linoleic (C18: 2) of 17.79%, linolenic (C18: 3) 1 , 21%, EPA 0.57%, and DHA 3.51%. Linoleic and linolenic fatty acids are essential fatty acids that cannot be produced by the body. Based on these data, catfish oil contains essential fatty acids which are quite high. MFish oil derived from fresh water (catfish, cork, and goldfish) can be used as a source of omega-6 fatty acids. In addition, oleic fatty acids (omega-9) also provide health benefits if consumed, including reducing cholesterol levels and preventing heart disease.. To increase the level of omega-3, it is necessary to add raw material for fish feed containing omega-3 based on the library, including micro algae, macroalgae, soybean oil or fish meal [2].

In the field of animal husbandry there has been research on omega-3 eggs on on chicken eggs or bird quail with low cholesterol and high omega-3 levels then conducted research omega-3 eggs with Omega-3 compositions are higher than normala. By learning the techniques of making omega-3 eggs, it will be reviewed how the technique of increasing catfish that are low in omega-3s becomes high, so that they become functional feeds that can increase the immunity of humans who consume them. The purpose of this review is the discovery of various candidatessources of raw materials which contains omega-3s which can be used to feed fish in order to increase the content of omega-3 fatty acids in fish.

Omega-3 Eggs

The use of lemuru fish oil in broiler rations apparently can reduce meat fat and cholesterol levels, but on the contrary reduce chicken performance. The use of both types of oil, both alone and in combination has been studiedin laying hens. The results show that the use of lemuru oil can reduce egg cholesterol content and increase Omega-3 content significantly, while the combination of the two types of oil (2% lemuru oil and 6% palm oil) provides egg yields with good egg production, quality, Omega-ratio 3, and Omega-6 in balanced eggs [3].

Th research has been done on quail (Coturnixcoturnix japonica) to determine the effect of lemuru oil and palm oil in rations both individually and in combination with the performance of quails and the content of Omega-3 fatty acids in their eggs. In addition, we want to know the most appropriate combination of the two oils in the feed to produce eggs with high Omega-3 content, low cholesterol levels without disturbing the performance of quails. Quail feed formulations were made with various levels of lemuru fish oil and palm oil as shown in Table 1.

Table 1. Feed Composition Quail

Treatment	Quail Laying Feed (%)	Concentrate (%)	Corn (%)	Rice bran (%)	Palm oil (%)	Lemuru Oil (%)	Crude protein (%)
Q1	100	0	0	0	0	0	21
Q2	0	50	30	20	0	0	21.1
Q3	0	50	30	20	8	0	21.1
Q4	0	50	30	20	2	6	21.1
Q5	0	50	30	20	4	4	21.1
Q6	0	50	30	20	2	6	21.1
Q7	0	50	30	20	0	8	21.1

Adopted from Suripta and Astuti (2007)

This shows that the addition of lemuru oil plays a role in reducing cholesterol content of quail eggs. The decrease is related to the increasing Omega-3 in rations containing lemuru oil can reduce egg cholesterol. This is consistent with the opinion of Parks et al. (1989) and Griffin (1992) that Omega-3 can inhibit cholesterol biosynthesis and reduce plasma VLDL-cholesterol and triglycerides. Factors that determine cholesterol content in eggs are egg yolk weight, small egg yolks contain lower cholesterol than large egg yolks. The Omega-3: Omega-6 ratio is best achieved by T7 followed by T6, T5 and T4, this shows the role of lemuru oil in increasing the omega 3 egg content [3].

In this study, it was seen that the higher the content of lemuru oil the smaller the cholesterol content of eggs. But it is not known whether the cholesterol content decreases in line with the decrease in the size of the yolk or indeed the cholesterol content decreases with the same size of the yolk. Omega-3 (ALA, EPA, DHA) content increased significantly with increasing lemuru oil, while the Omega-6 content jumped very high from only 1.62% in the control quail to 22.43% in T3 (quail with palm oil) and decreases with decreasing palm oil content, but remains high at T7, even without palm oil. Here it is seen that lemuru oil also affects the content of Omega-6 eggs, in addition to influencing the increase in Omega-3. Linoleic acid (Omega-6) in eggs decreases with increasing Omega-3 content, it is suspected that high Omega-3 will inhibit the synthesis of Omega-6, according to the opinion of Murray et al. (1995) that the biosynthesis of Omega-3 will inhibit the biosynthesis of Omega-6 by competing for the same enzyme system. The Omega-3 and Omega-6 ratios in eggs show eggs from feed containing palm oil and lemuru have a smaller ratio than controls [3].

Catfish (*Clarias Sp.*) as The Potential Fresh Water Fish Culture Species

Catfish (including one of the fisheries commodities that have good prospects to be developed because it can be cultivated in limited land and water sources with high stocking density, cultivation technology is relatively easy to be mastered by the community, its marketing is relatively easy and the business capital needed is relatively low [2]. Another advantage possessed is that catfish oil contains omega-3 fatty acids which are relatively high compared to other freshwater fish, but lower when compared to sea fish. The content of unsaturated fatty acids and omega-3 fatty acids EPA and DHA catfish oil might be influenced by the age and weight of fish. The highest omega-3 EPA fatty acid content (0.96%) was found in fish weighing 200-250 g / head and the lowest (0.84%) is found in fish weighing 100 g / head. The highest DHA content (3.14%) was found in fish weighing 100 g / head and the lowest (2.27%) is found in fish weighing 140 - 170 g / head.

Figure 1. Catfish

Candidates for Raw Materials Containing Omega-3

Microalgae

One source of marine biodiversity that contains fatty acids is microalgae. The content of fatty acids in microalgae such as diatoms consists of omega-3 unsaturated fatty acids EPA (eicosapentaenoic acid) and DHA (docosahexaenoic acid)[4]. Some of the micro algae that have been investigated for their fatty acid content are Chaetoseroscalcitrans, Chaetocerosgracilis, Skeletonemacostatum, and Thalassiosirapseudonana which have total fatty acids of 4.6-11.1%, CryptomonadChroomonassalina (10.9-11.9%) and Prymnesiophyte Pavlova lutheri (19.7%). Other Prymnesiophyte microalgae, Isochrysus sp., and green microalgae such as Nannochlorisatomus and Tetraselmissuecica have small amounts of fatty acids. Whereas in the group of green microalgae such as Dunaliellatertiolecta, no fatty acids were detected.

Table 2. Fatty Acid Contents in some microalgae

Microalgae	Linolenic Acid C18: 3n3	Eicosapentanoic Acid C20: 5n3	Docosahexanoic Acid C22: 6n3	Omega-3 total	Author
Pichochlorium Sp.	0.08	-	0.15	0.23	[5]
Isochrysisgalbana	3.8	0.8	15.8	20.4	[6]
Pavlova Sp	1.8	18.0	13.2	33.0	[6]
Phaeodactylumtricomutum	0.3	28.4	0.2	28.9	[6]
Rhodomonasbatica	12.0	4.4	-	16.4	[6]
Oocystis Sp.	8.1	1.1	-	9.2	[6]
Pseudokirchneriellasubscaptiata	11.4	-	0.1	11.5	[6]
Tetraselmis Sp.	6.4	4.8	0.2	11.4	[6]
Nannochloropsisoceanica	0.5	23.4	-	23.9	[6]
NannochloropsisSp	-	16.18	-	16.18	[7]

The microalga with a high content of omega-3 fatty acids shows inChroomonassalina, *Scenedesmus*sp. (5.6% moisture, 45.7% protein, 9.1% fat, 15.8% fiber and 8.3% ash). The study comprised three experimental diets: a control diet (CT) with a low level of fishmeal (10%) and relatively high levels of soy, pea and potato protein concentrates (1: 1: 1 blend), wheat gluten and corn gluten as major protein sources; a diet containing 10% Scenedesmus and 5% fishmeal (SCE10); and a diet with 20% Scenedesmus and 2.5% fishmeal (SCE20). The present study indicates that the incorporation of microalgae Scenedesmus sp. of up to 10% in low fishmeal diet did not affect the feed intake, growth and chemical composition of salmon. However, the inclusion of the microalgae, particularly at 20% in low fishmeal diets, significantly reduced the digestibility, nutrient retention efficiency and feed conversion ratio in Atlantic salmon. Scenedesmus sp. at 10% in the diet improved the total n-3 and PUFA content in salmon. The inclusion of the microalgae up to 10% also did not significantly alter the physical quality of the diet. [8]

The effects of the microalgae *Scenedesmusalmeriensis* incorporated to diets intended to feed sea bream (Sparusaurata) juveniles in a 45-day trial. Growth, body composition, and intestinal functionality of fish were studied. Microalgae meal was tested in triplicate at different inclusion levels (0%, 12%, 20%, 25% and 39%). At the end of the trial, individual body weight were recorded for evaluation of growth and nutrient utilization, and biological samples were obtained for proximate analysis, determination of digestive enzyme activities, intestinal histology, and microvilli morphological evaluation. Results indicated that S. almeriensis did not cause negative effects on fish growth or nutrient utilization efficiency. Growth rate and protein utilization in fish fed on a diet with 20% Scenedesmus meal tended to be higher,

Macroalgae

Omega-3 fatty acids that can be identified from the four types of seaweed are linolenic acid, echosatrienoicacid, EPA and DHA vary in amount. Fatty acid levels in the four types of seaweed ranged from 26.8% -52.26% and omega-3 content between 1.86% -5.46% [10].Macroalgae are a natural food source for many small aquaculture animals including shrimp larvae and fish seeds. Algal supplements contain various compounds that function as non-specific immunostimulants, increase innate defense mechanisms in animals, and provide increased resistance to pathogens. Development of algae genetic engineering technology enables the functioning of algae feed by algae cells to produce new bioactives, such as vaccines, growth hormones / anti-microbial agents. The species that are often used are Chlorella, Tetraselmis, Isochrysis, Pavlova, Phaeodactylum, Chaetoceros, Nannochloropsis, Skeletonema, and Thalassiosira[11].

Various macroalgae species exhibit natural anti-bacterial activity, and some also contain biomolecules that function as immunostimulants.Polyunsaturated fatty acids in long-chain algal derivatives such as eicosapentaenoic acid (EPA), as well as algal sterols, have anti-bacterial properties and can be effective against gram-positive and gram-negative bacteria. Substitution of fish oil with algae flour containing high amounts (DHA) and arachidonic acid (AA), significantly increases immune parameters [12].

Macroalgaeare knownhave a low lipid levels. However, compared to land-based vegetables they contain significantly higher levels of polyunsaturated fatty acids (PUFA) such as omega 3 and 6 fatty acids. PUFAs have reported antioxidant properties and play a crucial role in the prevention of cardiovascular diseases, osteoarthritis and diabetes[13] S. wightii lipids ranged from 2 to 3% [14]. *Sargassum* lipid content ranges from 1.5 to 3.5% in a variety of species [15, 16]

The most abundant fatty acids in E. cottonii was the omega-3 fatty acids, EPA which accounted for 24.98% of all fatty acids. *Sargassumpolycystum* contained a small quantity of docosahexaenoic acid. Palmitic (C16: 0) and oleic acids (C18: 1ω9) were the most abundant fatty acids in *C. lentillifera and S. polycystum, and E. cottonii* had the highest amount of omega-3 fatty acids (45.72%) compared to *S polycystum* (9.63%) and C. lentillifera (7.55%).

Table 3. Fatty acid composition of different species of macroalgae (% of total fatty acid content)

Components (ω3)	*Species*	*Composition (% of total fatty acid content)*	*Author*
α-Linolenic C18: 3ω3	*S. polycystum*	1.41	[17]
	*Sargassum*spp.	7.0; 7.2; 6.2; 6.3; 7.9; 6.7; 8.9	[18]
	E. cotonii	3.88	[17]
	Padinatetrastromatica	1.96	[19]
	Asperumspatoglossum	5.54	[19]
	Stoechospermummarginatum	9.95	[19]
	Cystoseiraindica	0.84	[19]
	Sargassumtenerrimum	0.96	[19]
	Kappaphycusalverzii	1.50	[19]
α-Linolenic C18: 4ω3	*Sargassum*spp.	7.4; 7.3; 7.5; 5.1; 6.5; 5.3; 7.5	[18]
Cis-11,14,17-Eicosatrienoic C20: 3ω3	*S. polycystum*	6.38	[17]
	*Sargassum*spp.	0.1; 0.3; 1.5; 1.2	[18]
	E. cotonii	16.87	[17]
	Padinatetrastromatica	1.07	[19]
	Asperumspatoglossum	00.42	[19]
	Stoechospermummarginatum	0.15	[19]
	Cystoseiraindica	0.06	[19]
	Sargassumtenerrimum	1.07	[19]
Cis-5,8,11,14,17-Eicosapentaenoic C20: 5ω3	*S. polycystum*	1.71	[17]
	*Sargassum*spp.	14.4; 3.8; 3.2; 4.6; 4.3; 4.5	[18]
	E. cotonii	24.98	[17]
	Padinatetrastromatica	3.51	[19]
	Asperumspatoglossum	11.7	[19]
	Stoechospermummarginatum	7.76	[19]
	Cystoseiraindica	7.28	[19]
	Sargassumtenerrimum	5.83	[19]
	Kappaphycusalverzii	11.1	[19]
Cis-4,7,10,13,16,19-Docosahexaenoic C22: 6ω3	*S. polycystum*	0.13	[17]
	Padinatetrastromatica	0.78	[19]
	Asperumspatoglossum	0.05	[19]
	Stoechospermummarginatum	0.29	[19]
	Cystoseiraindica	0.47	[19]
PUFAs ω3	*S. polycystum*	9.63 ± 0.15	[17]
	*Sargassum*spp.	19.7; 25.9; 22.0; 16.9; 20.8;	[18]
	E. cotonii	18.1; 20.5	[17]
	Padinatetrastromatica	45.72 ± 0.59	[19]
	Asperumspatoglossum	39.4	[19]
	Stoechospermummarginatum	49.6	[19]
	Cystoseiraindica	44.4	[19]
	Sargassumtenerrimum	47.5	[19]
	Kappaphycusalverzii	48.2	[19]
		27.0	[19]

Soybean Oil

Fish oil is a potential source of linoleic acid (Omega-6) and linolenic acid (Omega-3), but is relatively more difficult to obtain compared to vegetable oils. Vegetable oils derived from seeds contain relatively higher linolenic acid and linoleic acid compared to other vegetable oils such as coconut oil and palm oil. Grain oil containing linoleic acid and linolenic acid which is relatively higher and easily obtained is soybean oil[1].

Soybean oil given to catfish feed is expected to increase omega-3 EPA and DHA in catfish oil, also increase other omega-3 fatty acids such as linoleic acid and linolenic acid, while increasing unsaturated fatty acids of catfish oil. Increased use of soybean oil is very likely to increase the content of EPA and DHA of catfish oil, in the sense of a positive correlation between an increase in soybean oil content in feed with an increase in EPA and DHA content of catfish oil. However, there is also a chance of not having a positive correlation, having a pattern of change that follows the parabolic curve, meaning that there is a concentration that produces the optimum EPA and DHA.

Single and multiple unsaturated fatty acids including omega-3 fatty acids EPA and DHA which play a role in reducing levels of triacylglycerol and blood cholesterol levels as well as increasing the excretion process, increasing the fluidity of cell membranes, forming eicosanoids which reduce platelets and play an important role in brain and retinal development [1]. Omega-3 fatty acids are also able to prevent cardiovascular disease and improve the development of brain function and eye retina in infants [20]. With the broad spectrum of uses of omega-3 EPA and DHA fatty acids as well as easy cultivation of catfish, there needs to be efforts to increase the omega-3 EPA and DHA content of catfish oil. At present the availability of EPA and DHA is limited and expensive, this encourages us to look for alternative sources of freshwater fish, in this case cultured fish namely catfish. Some researchers have managed to improve the content of EPA and DHA fish oil through feeding containing omega-fatty acids3 [21]. Omega-3 fatty acids, linolenic acid and linoleic are found not only in fish oil, but also in vegetable oils, especially in grains. Soybean oil contains 49.4% linoleic acid and 9.1% linolenicacid[22].

The advantage of soybean oil is high content of linoleic acid and linolenic acid of soybean oil, and give a chance that soybean oil can be used to increase the content of omega-3 fatty acids EPA and DHA in catfish through feeding containing soybean oil. This is the background of this research. Catfish oil contains omega-3 fatty acids EPA and DHA which are relatively higher when compared with other freshwater fish, but still lower than seawater fish. Efforts to increase the content of omega-3 fatty acids EPA and DHA catfish are relatively easier compared to other freshwater fish, brackish water fish and sea water fish which are relatively difficult to cultivate. This increase can be achieved through the use of feed containing omega3 fatty acids EPA and DHA, can also be through the use of feed containing linoleic acid and high linolenic acid. This is because linoleic acid and linolenic acid are the precursors of EPA and DHA. Fish oil is one of the potential sources of linoleic acid and linolenic acid, but is relatively more difficult to obtain compared to vegetable oils. Vegetable oils derived from seeds contain linolenic acid and linoleic acid which are relatively higher compared to other vegetable oils such as coconut oil and palm oil. Grain oil containing linoleic acid and linolenicacid which is relatively higher and easily obtained is soybean oil. Soybean oil given to catfish feed is expected to increase omega-3 EPA and DHA in catfish oil, also increases other omega-3 fatty acids such as linoleic acid and linolenic acid, while increasing unsaturated fatty acids in catfish oil. Increased use of soybean oil is very likely to increase the content of EPA and DHA of catfish oil, in the sense of a positive correlation between an increase in soybean oil content in feed with an increase in the content of EPA and DHA of catfish oil. However, there are also opportunities not to have a positive correlation, have a pattern of change which follows a parabolic curve, in the sense that there is a concentration that produces the optimum EPA and DHA [1].

The Possibility Ways to Increase Omega-3 in Catfish

Catfish contain higher levels of omega-6 fatty acids compared with omega-3 fatty acids. The omega-3 fatty acid content of catfish is higher than that of other freshwater fish. Increasing the content of omega-3 fatty acids in catfish is relatively easier than other fresh fish, brackish fish or sea water fish. This increase can be done through the use of fish feed containing omega-3 fatty acids. Omega-3 fatty acids that can be added are EPA, DHA, and linolenic acid. P.feed bucket performed with certain formulations, namely the addition of soybean oil or sea fish oil as a source of fat.

The feed formulation provided is 25% soy flour, 10% fish meal, 60% fine bran, 5% tapioca and 10% soybean oil. Soybean oil feed mixture is given 3 times a day on catfish. Catfish fatty acid profile that was given supplementary formulations of soybean oil supplement were capric acid, lauric acid, myristic acid, palmitic acid, palmitoleic acid, stearic acid, oleic acid, linoleic acid, linolenic acid, behanic acid, arachidic acid, EPA and DHA .Feeding catfish containing soybean oil produces fish with high omega-3 levels. Catfish fed with an additional formulation of soybean oil 10% can increase omega-3 fatty acids 8.33%, EPA 5.05% and DHA 1.14%[1].

Conclusion

Single and multiple unsaturated fatty acids including omega-3 fatty acids EPA and DHA which play a role in reducing levels of triacylglycerol and blood cholesterol levels as well as increasing the excretion process, increasing the fluidity of cell membranes, forming eicosanoids which reduce platelets and play an important role in brain and retinal development. Omega-3 fatty acids are also able to prevent cardiovascular disease and improve the development of brain function and retinal eye in infants (Nettleton, 2005). EPA and DHA content of fish oil through feeding containing omega-3 fatty acids[21]. Omega-3 fatty acids, linolenic acid and linoleic are found not only in fish oil, but also in vegetable oils, especially in grains. Soybean oil contains 49.4% linoleic acid and 9.1% linolenicacid.With the high content of linoleic acid and linolenic acid of soybean oil, there is a chance that soybean oil can be used to increase the content of omega-3 fatty acids EPA and DHA in catfish through feeding containing soybean oil. This is the background of this research. Catfish oil contains omega-3 fatty acids EPA and DHA which are relatively higher when compared with other freshwater fish, but still lower than seawater fish. Efforts to increase the content of omega-3 fatty acids EPA and DHA catfish are relatively easier compared to other freshwater fish, brackish water fish and sea water fish that are relatively difficult to cultivate. This increase can be achieved through the use of feed containing omega-3 fatty acids EPA and DHA, can also be through the use of feed containing linoleic acid and high linolenic acid. This is because linoleic acid and linolenic acid are the precursors of EPA and DHA.

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