5. THE MOST POTENTIAL FISHBONE FLOURS AS AN ANTI-AGING INGREDIENT IN FOOD PRODUCT

Table 2.Nutrients in *Channa striata, Clarias batrachus*, and *Tilapia nilotica Fishbone*

Nutrients	***Channa Striata***	***Clarias batrachus***	***Tilapia nilotica***
Histidine	11.77 (1.17%) **(Rosmawati,2018)**	0% **(Shyni,2014)**	6 (0.6%) **(Haiying ,2015)**
Leucine	32.83 (3.28%) **(Rosmawati,2018)**	2.5% **(Shyni,2014)**	22 (2.2%) **(Haiying ,2015)**
Isoleucine	18.34 (1.83%) **(Rosmawati,2018)**	1.5% **(Shyni,2014)**	11 (1.1%) **(Haiying ,2015)**
Methionine	15.58 (1.55%) **(Rosmawati,2018)**	7.7% **(Shyni,2014)**	13 (1.3%) **(Haiying ,2015)**
Threonine	33.27 (3.32%) **(Rosmawati,2018)**	3.9% **(Shyni,2014)**	26 (2.6%) **(Haiying ,2015)**
Tyrosine	9.47 (0.94%) **(Rosmawati,2018)**	0.6% **(Shyni,2014)**	3 (0.3%) **(Haiying ,2015)**
Phenylalanine	30.76 (3.07%) **(Rosmawati,2018)**	2.9% **(Shyni,2014)**	13 (1.3%) **(Haiying ,2015)**
Trypthophan	0.95 (0.09%) **(Rosmawati,2018)**	**No study yet**	**No study yet**
Valine	27.98 (2.79%) **(Rosmawati,2018)**	2.5% **(Shyni,2014)**	21 (2.1%) **(Haiying ,2015)**
Lysine	54.24 (5.42%) **(Rosmawati,2018)**	3.5% **(Shyni,2014)**	26 (2.6%) **(Haiying ,2015)**
Aspartic Acid	59.49 (5.94%) **(Rosmawati,2018)**	6.5% **(Shyni,2014)**	44 (4.4%) **(Haiying ,2015)**
Glutamic Acid	115.29 (11.52%) **(Rosmawati,2018)**	9.1% **(Shyni,2014)**	75 (7.5%) **(Haiying ,2015)**
Alanin	99.95 (9.99%) **(Rosmawati,2018)**	9.1% **(Shyni,2014)**	119 (11.9%) **(Haiying ,2015)**
Arginin	81.53 (8.15%) **(Rosmawati,2018)**	6% **(Shyni,2014)**	51 (5.1%) **(Haiying ,2015)**
Serine	30.19 (3.01%) **(Rosmawati,2018)**	3.8% **(Shyni,2014)**	38 (3.8%) **(Haiying ,2015)**
Cysteine	0.33 (0.03%) **(Rosmawati,2018)**	0% **(Shyni,2014)**	0% **(Haiying ,2015)**
Glycine	235.25 (23.5%) **(Rosmawati,2018)**	21.2% **(Shyni,2014)**	33.7% **(Haiying ,2015)**
Proline	121.81 (12.1%) **(Rosmawati,2018)**	9% **(Shyni,2014)**	11% **(Haiying ,2015)**
Hydroxyproline	11.13% **(Rosmawati,2018)**	5.3% **(Shyni,2014)**	7.6% **(Haiying ,2015)**
Moisture	43.19% **(Rosmawati,2018)**	11.43% **(Shyni,2014)**	14.2% **(Trilaksani *et al,* 2006)**
Protein	15.49% **(Rosmawati,2018)**	24.3% **(Sa'adah, 2013)**	40.8% **(Trilaksani *et al,* 2006)**

.....Table 2

Lipid	4.19% **(Rosmawati,2018)**	3.84% **(Sa'adah, 2013)**	25.3% **(Trilaksani *et al,* 2006)**
Ash	32.05% **(Rosmawati,2018)**	58.43% **(Sa'adah, 2013)**	18.3% **(Trilaksani *et al,* 2006)**
Collagen	33% **(Liu et al , 2009)**	16.4% **(Normah,2012)**	40.8x35% = 14.28% **(Rehn *et al*,2001)**
Carbohydrate	5.08% **(By Difference Calculation)**	6.02% **(Sa'adah, 2013)**	1.4% **(By Difference Calculation)**
Calcium	39% **(Liu et al , 2009)** 1406mg/100g (1.4%)-1949mg/100g (1.95%) **(Ryo,2015)**	6.33% **(Bechtel,2019)** & 0.735% **(Sa'adah, 2013)**	2715.9mg/100g (2.72%) **(Trilaksani *et al,* 2006)** 52085mg/kg (5.2%) **(Kasozi,2018)**
Potassium	0.2% **(Liu et al , 2009)**	0.51% **(Bechtel,2019)**	9450 mg/kg (0.945%) **(Kasozi,2018)**
Sodium	0.7% **(Liu et al , 2009)** 1.7 mg/g (0.17%) **(Tawali,2019)**	0.27%**(Bechtel,2019)**	**No study yet**
Magnesium	0.5% **(Liu et al , 2009)** 1.51 (0.15%) mg/g **(Tawali,2019)**	0.13%**(Bechtel,2019)**	**No study yet**
Carbonate	9.8% **(Liu et al , 2009)**	**No study yet**	**No study yet**
Phosphate	17% **(Liu et al , 2009)**	**No study yet**	**No study yet**
Phosphorus	0.693 mg/g **(Tawali,2019)**	3.27%**(Bechtel,2019)**	1132.7mg/100g (1.13%) **(Trilaksani *et al,* 2006)** 30554 mg/kg (3.06%) **(Kasozi,2018)**
Copper	0 mg/g **(Tawali,2019)**	<0.2 ppm (<0.0002 mg/g) **(Bechtel,2019)**	**No study yet**
Zinc	0.0585 mg/g (0.00585%) **(Tawali,2019)**	48.87 ppm (0.004887%) **(Bechtel,2019)**	217.2 mg/kg (0.2172%) **(Kasozi,2018)**
Manganese	**No study yet**	8.89 ppm (0.00889 mg/g) **(Bechtel,2019)**	12.3 mg/kg (0.00123%) **(Kasozi,2018)**
Nickel	**No study yet**	<0.2 ppm (<0.0002 mg/g) **(Bechtel,2019)**	**No study yet**
Iron	0.046 mg/g **(Tawali,2019)**	6.16 ppm (0.00616 mg/g) **(Bechtel,2019)**	1.3mg/100g (0.0013%) **(Trilaksani *et al,* 2006)** 15.1 mg/kg (0.0015%) **(Kasozi,2018)**

5.1.Essential Amino Acids

Histidine is a precursor to histamine (a neurotransmitter that plays a role in the control of food intake, along with other neurotransmitters) that help to build protein and has an anti-inflammatory role to help ease joint pain and stiffness, also has antifungal functions, can decrease fatigue, increases concentration and efficiency at work, decrease severity in atopic dermatitis disease, and can induced anorexia (Moro, 2020). Leucine is an essential branched-chain amino acid that helps increase muscle mass and help the recovery of muscle after exercise, regulates blood sugar, supply the body with energy, promotes energy metabolism (glucose uptake, mitochondrial biogenesis, and fatty acid oxidation), help the body to heal, and affects brain functions (Duan, 2015). Isoleucine is important in hemoglobin synthesis and its primary function is to boost up the energy levels and assist the body recovering from strenuous physical activity, so it is recommended to athletes (Jojima, 2010).

Methionine is an aliphatic, sulfur-containing, and essential amino acid, also a precursor of succinyl-COA, homocysteine, cysteine, creatine, and carnitine that used for increasing the acidity of urine, treating liver disorders, improving wound healing, treating depression, alcoholism, allergies, asthma, copper poisoning, radiation side effects, schizophrenia, drug withdrawal, and Parkinson's disease (Martinez, 2017). Threonine is a nutrient that plays a critical role in the maintenance of intestinal mucosal integrity and barrier function that also helps in the synthesis of glycine and serine which help in the production of collagen, elastin, and muscle tissue. It also supports cardiovascular, liver, central nervous, immune system functions and the growth of thymus, aids in maintaining strong bone and tooth, speed up wound healing, reduces the accumulation of fat in the liver, and useful in treating Lou Gehrig's disease (Amyotrophic Lateral Sclerosis or Motor Neurone Disease) (Mao,2011). Tyrosine is the precursor of the catecholamines that can influence the synthesis of both dopamine and norepinephrine in the brain and also can help to suppress appetite, promote weight loss, increase mental alertness, improve memory, enhance athletic performance, and can treat phenylketonuria (a genetic disorder) (Young, 2007).

Phenylalanine is an amino acid that was strongly associated with mortality by helping in reduce mild pain symptoms, joint discomfort, and even out skin tone because it promotes balanced neural and cognitive function, also plays role in regulating mood (Huang, 2019). Tryptophan is an important natural mood-lifting substance that gets converted into serotonin (the happy hormone) which can encourage weight loss and help overcome sleep disorder (Kaluzna, 2019).

Valine is a branched-chain amino acid associated with tissue repair, muscle metabolism, muscle co-ordination, mental vigor, and calm emotion (Nagao, 2012). Lysine is an essential cationic amino acid that has a positively charged R group which can improve athletic performance, also preventing and treating cold sores. Lysine has a role in cellular proliferation, the ability to induce strong inflammatory and immune response (humoral and cell-mediated), augmented healing of all types of wounds, and inducing extensive angiogenic responses (Datta, 2001). Table 2 showed that *Channa striata* bones have the highest histidine, leucine, isoleucine, tyrosine, phenylalanine, valine, and lysine contents than other fishes. While methionine and threonine are highest in *Clarias batrachus* bones than in snakehead and nile tilapia. Due to the limitation of the study, The data of tryptophan content in this study can not be used as a factor to study the most potential fishbone flour as an anti-aging ingredient. The result of this study is uncertain and the degree of error is quite high because the data obtained from various literatures which not under the same standard. A further experiment under the same standard is needed to see the most potential fishbone for an anti-aging ingredient in food products with minimum experimental error and more certain data.

5.2.Non-Essential Amino Acids

Aspartic acid or aspartate is a non-essential amino acid which is chiral and exist in two enantiomeric forms that important to the development of the nervous system, enhancing secondary calcium-dependent neurotransmission signaling, decreasing risk factors of cardiovascular disorders, increasing testosterone levels, help to gain lean muscles mass, decrease fat stores, stimulate the increase in muscle power, promote hormone synthesis, increasing fertility, and enhancing spermatogenesis (Johnson, 2017). Glutamic acid helps in protein synthesis also has several key functions within the body and is critical for healthy brain development and function. It also considered protective against obesity and cardiovascular disease (Takahashi, 2011). Alanine is the principal amino acid released by the forearm muscle of man and is the principal amino acid extracted by the liver for gluconeogenesis which is important to maintain glucose and nitrogen levels that make energy is supplied in the body. Alanine also helps to protect cells from being damaged during intense aerobic activity (when the body cannibalizes muscle protein to help energy production) (Felig, 1970).

Arginine is an anti-inflammatory that fights migraines, help phosphorus and calcium build bones and teeth, and is an essential component for spermatogenesis, embryonic growth, fetal

and neo fetal growth, also for immune and tissue integrity (Wu, 2009). Glycine is the smallest non-essential, neutral, and metabolically inert amino acid with a carbon atom bound to two hydrogen atoms and to an amino and a carboxyl group which is a content that is foundational to DNA and RNA strands creation. DNA and RNA can block endotoxin and help to transport nutrients for body cells to utilize energy. Glycine in fish collagen is known as the anti-aging amino acid that has a broad spectrum of anti-inflammatory, cytoprotective, and immunomodulatory properties that also help to stabilize blood sugar, form new muscle, aids in muscle recovery, and prevent cartilage loss. It also participates in the synthesis of proteins, tripeptide glutathione, and in detoxification reactions (Torres, 2016). Proline is a non-essential amino acid, and along with hydroxyproline is the only amino acid without a primary amino group, therefore has a unique metabolic pathway in the mitochondria that is foundational to the human body's ability to naturally produce its collagen by helping the stimulation of the process within the body. Proline also acts as an antioxidant for the body and prevent cell damage from free radicals. The skin with additional proline will have more elastic properties and become a healthy age-defying skin (DeArmond, 2017).

Serine is a non-essential amino acid directly involved in cellular homeostasis, proliferation, and differentiation that also a constituent in the brain and also a protective cover of nerves that helps to synthesis protein, fatty acid, genetic code carriers such as DNA and RNA, also help in the building of muscles. It is an important amino acid for brain and nervous system to work properly, enhance retinal health, and boosts a healthy immune system by aiding the production of antibodies (Sinha, 2020). Cysteine is a protogenic amino acid that has positive roles in the immune system, skin and hair, male fertility, inflammation, osteoporosis, anti-aging, and general well-being of the body. Cysteine is classified as semi-essential amino acid and can be synthesized in the liver from methionine which is a sulfur-containing amino acid (Sameem, 2018). Hydroxyproline plays a critical role in stabilizing the triple helix of collagen and is suggested to be a candidate compound for treatment or reduction of wrinkles because the triple-helical conformation gives collagen most of its unique properties and is essential for normal fibrillogenesis (Suh, 2007).

Hydroxyproline is the digestive products of collagen hydrolysate which is known as collagen peptide that suggested to improved aged or damaged skins (Lee, 2008). These bioactive peptides have antioxidant properties that held the reduction of reactive oxygen species which are unstable molecules containing oxygen that cause the aging of tissues and lead to a higher

risk of cancer also it can boost the immune system and reduce inflammatory responses throughout the body (Aleman, 2011). Bioactive peptides found within fish collagen also have antibacterial properties which particularly consist of peptide collagencin that helps inhibit the growth of infections and bacteria which cause disease (Ennaas, 2016). Table 2 showed that *Channa striata* bones have the highest glutamic acid, arginine, cysteine, proline, and hydroxyproline contents than other fishbones. *Clarias batrachus* bones have the highest aspartic acid and serine contents, while *Tilapia nilotica* bones have the highest alanine and glycine contents than other fishbones. Hydroxyproline content in fishbone flours can make food products have better thermostability, rheological properties, and gel strength if it has been added. Due to the hydroxyproline content, Food products of *Channa striata* fishbone flour are to be estimated to have the best gel strength, thermostability, and rheological properties than other food products from *Clarias batrachus* or *Tilapia nilotica* fishbone flours.

5.3.Macronutrients and Mineral

Protein is the major functional and structural component of all the cells in the body. Enzymes, membrane carriers, blood transport molecules, the intracellular matrices, hair, fingernails, serum albumin, keratin, and collagen are proteins. The constituent amino acids of protein act as precursors of many coenzymes, hormones, RNA, DNA, and other essential molecules for life. An adequate supply of protein is essential to maintain health, reproduction, and cellular integrity and function (Food and Nutrition Board, 2005). Proteins in fish are highly digestible compared to other terrestrial animals and plants. Fish protein will break down on digestion into polypeptides, peptides, and amino acids that have bioactive properties. Table 2 showed that nile tilapia has the highest protein content in its bones and for the lowest one is snakehead fish. Carbohydrate is also a macronutrient that present in good amounts in fish, so it will give energy and structural components with monosaccharides, disaccharides, and polysaccharides. Collagen is known as the most abundant protein in vertebrates and contributes significantly to the hardness of connective tissues such as skin, bones, cartilage, tendons, and blood vessels (Liu, 2012). Due to its biological compatibility, biodegradability, low cytotoxicity, structural support, and hemostatic activity, Collagen is widely used in several industries including the food industry (Schmidt, 2016). If added in food products, Collagen can help to inhibit the denaturation of myofibril protein that enhace Water Holding Capacity in food product. Table 2 showed that the highest carbohydrate content is found in *Clarias batrachus* fishbone. The highest collagen content is found in *Channa striata* fishbone. While *Tilapia nilotica* fishbone

has the lowest content in both carbohydrate and collagen. Based on the collagen content, Food Products made of *Channa striata* fishbone flours are to be estimated have higher Water Holding Capacity than those made from *Clarias batrachus* or *Tilapia nilotica* fishbone flours.

Calcium is the most common mineral in the human body because 99% of it is found in bones and soft tissue of the human body. If the calcium levels in the blood are inadequate, the body will demineralize bone to maintain the calcium content (Heaney, 2000). Calcium plays a role in mediating blood vessels relaxation, nerve impulse transmission, muscle contraction, enzyme activation, membrane permeability, blood clotting, and the secretion of hormones like insulin (Food and Nutrition Board,1997). The deficiency of calcium can cause rickets in children, osteomalacia, and osteoporosis in adults also affects the dentition of both children and adults. (Hays, 1985). Several studies in Table 2 have shown a contradictory relationship to the calcium content in the bones of the same fish. This contradictory relationship is shown in the range of calcium content which is too wide in the same fishbone. Therefore, The data of calcium content in this study can not use to be a factor that affects the goals of this study.

Phosphorus is an essential mineral that is required by every cell in the body and 85% of it is found in the bone of the human body (Food and Nutrition Board,1997). Phosphorus is a constituent of bones, teeth, adenosine triphosphate (ATP), phosphorylated metabolic intermediates, and nucleic acids (Murray, 2000). The activation of some enzymes, hormones, and cell-signaling molecules depends on phosphorylation. Deficiency of phosphorus can lead to rickets, hyperparathyroidism, osteomalacia, chronic nephritis, and De Toni-Fanconi Syndrome (Malhotra, 1998).

Potassium is an essential dietary mineral and electrolyte (Peterson, 1997) that involved in the maintenance of membrane potential and as a co-factor for several enzymes (Sheng, 2000). An inadequate level of potassium may lead to hypokalemia that may result in muscular paralysis or abnormal heart rhythms (cardiac arrhythmias) if become severe (Sheng, 2000).

Sodium is the principal cation in extra-cellular fluids that regulates plasma volume and acid-base balance, involved in the maintenance of osmotic pressure of body fluids, preserves normal irritability of muscles and cell permeability, activates nerve and muscle function, and involved in sodium or potassium- ATPase maintenance of membrane potentials, the transmission of nerve impulse and the absorptive processes of monosaccharides, amino acids, pyrimidine and

biles salts (Murray, 2000). Magnesium is a mineral that plays important roles in the structure and the function of the human body because it involved in more than 300 essential metabolic reactions (Spencer,1994) such as energy production, synthesis of essential molecules, structural roles, ion transport across cell membranes, cell signaling, cell migration, *etc*. (Rude, 2006).

Copper is a constituent of many enzymes that plays a role in the absorption of iron (Chandra,1990). It is also an essential micro-nutrient which is necessary for the hematologic and neurologic systems (Tan, 2006), the growth and formation of bone, the formation of myelin sheaths in the nervous systems, helps in the incorporation of iron in hemoglobin and the absorption of iron from the gastrointestinal tract (GIT), and the transfer of iron from tissues to the plasma (Murray,2000).

Manganese plays important roles in some physiologic processes as a constituent of bone enzymes and an activator of other enzymes (Nielsen,1999). Manganese is involved in antioxidant function, metabolism, bone development, and wound healing because it is a co-factor of hydrolase decarboxylase and transferase enzymes (Murray,2000). Inadequate levels of manganese can cause skeleton deformities and defects in shell quality (Gordon,1997). Even it is found in small amounts in the human body, it plays a part in our overall health and bodily processes.

Nickel may help as a critical cofactor to enzymes to speed up the normal chemical reactions in the body. It is found in human RNA and DNA, where it interacts with these nucleic acids. But excess nickel will become toxic and can have severe side effects on the body (Nielsen,1999).

Iron is a key element in the metabolism of organisms (Fairbanks,1999) and is required for many vital functions, including growth, healing, and immune function. Deficiency of Iron can alter many metabolic processes such as neurotransmitter metabolism, protein synthesis, organogenesis, also may impact in brain function (Beard,1997). Zinc plays an important role in growth and development, the immune response, neurological function, and reproduction. Zinc function on the cellular level can be divided into three categories which are catalytic, structural, and regulatory (Cousins, 2006). There are about 100 different enzymes that depend on zinc for their ability to catalyze vital chemical reactions (Food and Nutrition Board,1997). Zinc has roles in cell replication, gene expression, Vitamin A and E metabolism and

bioavailability, also in nucleic acid and amino acid metabolism (Szabo,1999). This mineral is required for tissue repair and wound healing, plays vital roles in protein synthesis and digestion, necessary in optimizing insulin action, and acts as an important constituent of plasma (Malhotra,1998). Based on 12 minerals that have been found in this study , 8 of them (calcium, sodium, magnesium, carbonate, phosphate, copper, manganase, and nickel) can not be used as factors to study the potential of fishbone as an anti-aging ingredient due to limitation of the study. Table 2 showed that from 4 minerals that can be compared, 3 of them which are Iron, Potassium, and Zinc are highest in *Tilapia nilotica* fishbone while Phosphorus is highest in *Clarias batrachus* fishbone. Further experiment under the same condition is necessary to get accurate and complete data of minerals content in these fishbones.

5.4.Fatty Acids

Table 3.Fatty Acids in *Channa striata*,*Clarias batrachus*,and *Tilapia nilotica*

Fatty Acids	***Channa Striata***	***Clarias batrachus***	***Tilapia nilotica***
Palmitic Acid (16:0)	21.86%	24.34%	20.80%
Oleic Acid (18:1(N-9))	39.26%	38.18%	28.45%
Linoleic Acid (18:2(N-6))	19.58%	18.86%	20.55%
Linolenic Acid - LNA (18:3(n-3))	0.54%	0.78%	1.67%
Eicosapentaenoic Acid - EPA (20:5(n-3))	0.13%	0.05%	0.35%
Docosahexaenoic Acid - DHA (22:6(n-3))	0.68%	0.27%	4.66%
Saturated Fatty Acid (SFA)	29.68%	32.89%	31.58%
Monounsaturated Fatty Acid (MUFA)	45.45%	43.61%	34.13%
Polyunsaturated Fatty Acid (PUFA)	24.95%	23.19%	33.79%

Source: (Ghassem,2009)

There is a limitation in the study about fatty acids content in these fishbones. Only a study about fatty acids content in *Tilapia nilotica* fishbones that was found. There is no study of fatty acids content in the fishbone of *Channa striata* or *Clarias batrachus*. Fortunately, there is a literature that discuss about fatty acids content in these three fishes under the same condition. The data from that literature was used in this study to compares fatty acids content in these three fishes as factors to see the most potential fish as an anti-aging ingredient in food products. Palmitic acid (16:0) is the most common saturated fatty acid found in the human body and can be provided in the diet or synthesized endogenously from other fatty acids, carbohydrates, and amino acids that represent 20–30% of total fatty acids (FA) in membrane phospholipids (PL),

and adipose triacylglycerols (TAG) (Carta *et al*., 2015). A balance amount of Palmitic acid in the body can aid in membrane protein-lipid composition, lung surfactant superficial tension, proteins palmitoylation, and become the precursor of palmitoylethanolamide (PEA). However in unbalance condition, Palmitic Acid can cause tumor growth and invasiveness, inflammation, metabolic disorders, and Non-Alcoholic Fatty Liver Disease (NAFLD) (Carta,2017).

Saturated Fatty Acid (SFA) is the total amount of all saturated fats that is a major risk factor in hypercholesterolemia and cardiovascular disease if its amount is excessive in the human body. Lauric acid, Myristic acid, and Palmitic acid are the three principal cholesterol-raising effect of all saturated fatty acids. The critical level of cholesterol intake each day is 400 mg per day (Khosla,1994). Table 3 showed that *Clarias batrachus* has the highest content of palmitic acid and total saturated fatty acids than other fishes.

Oleic acid [C18:1(n-9)] is a monounsaturated fatty acid that mainly found in *the cis* form in nature. It has some beneficial health effects to human which contribute to the maintenance of normal blood cholesterol levels, modulation of inflammatory markers, blood pressure, insulin sensitivity, gastrointestinal functions, help in preventing the progression of metastasis in several human cancers and reduce the risk of developing certain types of cancer especially colorectal, breast, and prostate cancer (Pravst,2014). Monounsaturated Fatty Acid (MUFA) is the total amount of monounsaturated fats that can reduce traditional risk factors defining metabolic syndrome (MetS) and CVD. The consumption of dietary MUFA can promote healthy blood lipid profiles, mediates blood pressure, improve insulin sensitivity, influencing body composition, ameliorating the risk of obesity, and regulates glucose levels (Gillingham,2011). Table 3 showed that the highest oleic acid and total monounsaturated fatty acid contents are found in *Channa striata*.

Linoleic Acid (18:2(N-6)) is the most highly consumed PUFA found in the human diet and is an essential (indispensable) nutrient that contains 2 double bonds at the ninth and 12th carbons from the carbonyl function group. These fatty acids cannot be synthesized so it must be consumed through the human diet. Linoleic acid is involved in the maintenance of the transdermal water barrier of the epidermis (Whelan, 2013). This fatty acid is also an aid in lowering blood cholesterol levels, has effects on cardiovascular health, and has a role in slowing the development of atherosclerosis (Jandacek, 2017). Linolenic Acid - LNA (18:3(n-3)) is known as an omega-3 fatty acid which is an essential fatty acid that cannot be synthesized

by the body and must be supplied by dietary sources. Linolenic acid is known to have health benefits because it may play an important role in many physiological functions. These potential benefits of linolenic acid are cardioprotective effects, modulation of the inflammatory response, and a positive impact on both central nervous system function and behavior (Stark, 2008).

Eicosapentaenoic acid - EPA (20:5 (n-3)) and Docosahexaenoic Acid - DHA (22:6(n-3)) are long-chain omega-3 polyunsaturated fatty acids that can reduce the risks of cardiovascular disease (CVD) such as myocardial infarction, stroke, coronary artery disease, and sudden cardiac death. These two fatty acids also play important role in reducting the risk of cancer, Alzheimer's disease, depression, schizophrenia, fetal development particularly neuronal and retinal functions, and reduce the risks of various chronic diseases (Mateos, 2013). According to Duffy (2004), consumption of fish oil that rich of EPA can help patients to cure lupus which is a chronic inflammatory disease with the symptoms of photosensitive facial rash, fatigue, anorexia, weight loss, and sweating. This disease can threaten the heart, lungs, kidney, and central nervous system. Supplementation of omega-3 fatty acids especially EPA also can inhibit the formation of rheumatoid arthritis inflammatory agents (Kremer,1987), can dampen the adverse effects of leukotriene (metabolites of arachidonic acid) and has significant anti-inflammatory effects that use to treat psoriasis (a common disease characterized by a red inflamed border) (Bittiner,1988).

Fish oil also can help to cure Crohn's disease (an inflammatory disease involving intestinal pains, diarrhea, and malabsorption of the nutrients) (Tsujikawa, 2000), alleviate ulcerative colitis (Alex,1992), helps in lung and respiratory disorders like asthma (Nagakura, 2000), and kidney disorders (Donadio,1999). Omega-3 fatty acids especially DHA are positively associated with bone mineral accrual and with peak bone in young men that potential to the attainment of peak bone mass in adolescence and prevent age-related osteoporosis (Hogstrom,2007). DHA is abundant in the retina and important for visual acuity that prevents macular degeneration and has an influence on the development of visual impairment and improves visual function in the elderly (Simopoulos,1999). Omega-3 from fish oil is an important ingredient for the beauty and health of skin, hair, and nail (Sarojnalini, 2019).

Polyunsaturated Fatty Acid (PUFA) is a fatty acid that has the potential to be beneficial to health because it contains more than one unsaturation in their feature. PUFA has beneficial impacts on chronic diseases, cardiovascular disease, cancer, influence on gene expression,

immunological system, inflammation, pregnancy, breastfeeding, and infant development (Kus-Yamashita,2016). Table 3 showed that the highest content of linoleic acid, LNA, EPA, DHA, and polyunsaturated fatty acids are found in *Tilapia nilotica*.

Table 4.Comparison of Fatty Acids in *Tilapia nilotica* Bones and *Tilapia nilotica*

Fatty Acids	***Tilapia nilotica* Bones (Maria,2008)**	***Tilapia nilotica* (Ghassem,2009)**
Palmitic Acid (16:0)	208.5 mg/100g (0.208%)	20.80%
Oleic Acid (18:1(N-9))	344.3 mg/100g (0.344%)	28.45%
Linoleic Acid (18:2(N-6))	109.6 mg/100g (0.109%)	20.55%
Linolenic Acid - LNA (18:3(n-3))	29.9 mg/100g (0.0299%)	1.67%
Eicosapentaenoic Acid - EPA (20:5(n-3))	3.3 mg/100g (0.0033%)	0.35%
Docosahexaenoic Acid - DHA (22:6(n-3))	12.9 mg/100g (0.0129%)	4.66%
Saturated Fatty Acid (SFA)	296.2 mg/100g (0.296%)	31.58%
Monounsaturated Fatty Acid (MUFA)	4415.0 mg/100g (4.415%)	34.13%
Polyunsaturated Fatty Acid (PUFA)	175.6 mg/100g (0.1756%)	33.79%

Table 4 showed the comparison between fatty acids content in *Tilapia nilotica* bones and *Tilapia nilotica* in the fish form. The data in Table 4. can be used in the calculation to see the ratio of fatty acids content in fish and its bones. The calculation process has shown that the amount of all fatty acids in fishbone are lesser than in fish with different ratios on each type of fatty acids. The palmitic acid content in Nile tilapia bones is 100 times lesser than in fish, oleic acid is 83 times lesser, linoleic acid is 189 times lesser, LNA is 56 times lesser, EPA is 106 times lesser, DHA is 361 times lesser, total SFA is 107 times lesser, MUFA is 8 times lesser, and PUFA in fishbone is 192 times lesser than Nile tilapia fish. The fatty acids content in Nile tilapia bones is ranging from 8 to 361 times lesser than in fish. Further experiment under the same condition is needed to get accurate data of fatty acids content in fishbone flours.