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REVIEW ARTICLE

Omega 3-Induced Autophagy: A Two-Sided Sword in Cell Death from Historical Perspective in Clinical Application and Biological Mechanisms

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ABSTRACT

Omega-3 fatty acids, including eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA), and α-linolenic acid (ALA), are polyunsaturated fatty acids (PUFAs) with numerous health benefits. They play a critical role in various cellular functions, such as maintaining cell membrane integrity, cellular signaling pathways, and cell-to-cell communication. Omega-3s are known for their anti-inflammatory, anti-cancer, anti-depressant, and lipid-lowering properties. Autophagy, a cellular process responsible for recycling and degrading cellular components, is closely linked to these fatty acids. The impact of omega-3 fatty acids on autophagy signaling pathways hold promise for innovative therapeutic strategies in disease management and overall well-being. This review explores the influence of omega-3 fatty acids on autophagy and provides insights into the potential mechanisms involved.

Introduction

Nutrient intake pattern and lifestyle can greatly affect health status and cellular function (1). Among nutritional ingredients, omega-3 fatty acids were shown to be essential components of the broader category of polyunsaturated fatty acids (PUFAs) (2). They are classified into three main types of eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA), and α-linolenic acid (ALA) (3). These PUFAs, embedded in membrane phospholipids, play a crucial role in several cellular functions, including maintaining cell membrane structure and fluidity, facilitating cellular signaling pathways, and enabling cell-to-cell interaction (4). Omega-3s exhibit anti-inflammatory, anti-cancer, antidepressant, triglyceride- lowering, and low-density lipoprotein (LDL)-reducing properties (5-8). These properties are mediated through various mechanisms, including apoptosis, autophagy, and cell cycle regulation (8).

Autophagy is a catabolic process that maintains cellular homeostasis by recycling or degrading cytoplasmic components (9, 10). In 2016, The Nobel Prize in the field of Medicine was conferred

in recognition of the elucidation of the molecular mechanisms governing autophagy (11, 12). To date, three distinct forms of autophagy have been identified including macroautophagy, microautophagy and chaperone-mediated autophagy (13). Autophagy is stimulated by various triggers such as hypoxia, starvation, oxidative stress, and toxic molecules to decrease stress (9).

The initiation of autophagy involves the creation of double-membrane vesicles known as autophagosomes. These autophagosomes engulf the targeted materials and deliver them to lysosomes for breakdown and recycling. Autophagy signaling is regulated by various stimulators and inhibitors. Protein kinase B (PKB-AKT) pathway can activate Mechanistic Target of Rapamycin (mTOR) and inhibit autophagy. Conversely, the activation of autophagy is prompted by AMP-activated protein kinase (AMPK) (9). Numerous studies have reported that dietary intake or supplementation of omega-3 PUFAs to be associated with enhanced autophagy. Reactive oxygen species (ROS) accumulation induced by DHA is responsible for the Akt-mTOR signaling inactivation; thus, autophagy is induced (14). On the other hand, omega-3 may activate Akt-mTOR pathway to inhibit autophagy in some situations (15).

The dysregulation of autophagy may exert a highly significant influence on a diverse range of diseases. There is a clear correlation between the activation of autophagy-promoting molecules and the suppression of tumor formation, and similarly, molecules that impede autophagy that are linked to the development of cancer. It seems that autophagy offers protection against a range of neurodegenerative diseases. Impaired autophagy could potentially contribute to cardiac dysfunction (16, 17). This narrative review summarizes the current experimental evidence regarding the role of omega-3 fatty acids in autophagy signaling, focusing on potential mechanisms.

Nutritional Aspects: Nutritional Properties

Based on current knowledge, omega-3 fatty acids exhibit beneficial nutritional properties that promote health and reduce the risk of various diseases (18). Omega-3 fatty acids are associated with numerous nutritional effects, including anti-inflammatory, antioxidant, anti-proliferative, and anti-angiogenic properties (19, 20). Their triglycerides (TG)-lowering, antithrombotic, anti-inflammatory, and antihypertensive effects make them particularly valuable in the prevention and management of cardiovascular diseases (21, 22). Several epidemiological studies have demonstrated

the antidepressant effects of omega-3 fatty acids. Additionally, omega-3 fatty acids exhibit anti-carcinogenic properties and may enhance the efficacy and tolerability of chemotherapy (18). It was suggested that these fatty acids may be useful in obesity management by reducing appetite and modulating the expression of key genes involved in energy metabolism (23). However, high doses of omega-3 fatty acids may lead to hyperglycemia by enhancing glycerol gluconeogenesis (24).

Metabolism

Omega-3 fatty acids are classified within the PUFAs category. Since ALA cannot be synthesized by animals, it is considered an essential fatty acid. ALA, the most fundamental member of biologically active fatty acids, can undergo desaturation and elongation to convert into other omega-3 PUFAs. Although EPA and DHA can originate from ALA, the conversion process is highly limited. The genes encoding the desaturation enzymes are identified as fatty acid desaturase 1 and 2 (FADS1 and FADS2). Polymorphisms in these genes can lead to variations in the activity of desaturase enzymes (18, 25, 26).

EPA and DHA are precursors for eicosanoids such as prostaglandins (PGs), prostacyclin (PGI), thromboxane (TX), leukotrienes, hydroperoxy tetraenoic acid, hydroxyeicosatetraenoic acid, and lipoxins which participated in several physiological functions (18). Eicosanoids are formed through three primary enzymatic pathways of cyclooxygenase (COX), lipoxygenase (LOX), and cytochrome P450s (CYP) (27). Eicosanoids play a crucial role in inflammatory processes. Arachidonic acid-derived eicosanoids act in a pro-inflammatory way, while EPA-derived eicosanoids may oppose the function of those produced from arachidonic acid (26). Prostaglandins and leukotrienes derived from omega-3 PUFAs exhibit anti-inflammatory functions (28). EPA and DHA synthesize resolvins which have anti-inflammatory property (26).

Structural Features and Sources

Omega-3 fatty acids constitute a category of fatty acids characterized by a double bond between the third and fourth carbon atoms from the methyl terminal of the fatty acid chain. This structural feature leads to their designation as omega-3 fatty acids, n-3 fatty acids, or ω-3 fatty acids (n-3 FAs) (18, 29). ALA (cis-9, cis-12, cis-15-octadecatrienoic acid, 18:3) elongates and desaturates to convert to EPA (cis-5, cis-8, cis-11, cis-14, cis-17-eicosapentaenoic acid, 20:5) or DPA (cis-7, cis-10, cis-13, cis-16, cis-19-docosapentaenoic acid, 22:5) (18). DPA produces DHA (cis-4, cis-7, cis-10, cis-13, cis-16, cis-19-

docosahexaenoic acid, 22:6) as an intermediate fatty acid between EPA and DHA (30). The main sources of EPA and DHA in the diet are marine organisms particularly from the liver of lean white fish like cod and halibut, and the bodies of oily fish like menhaden, mackerel, and salmon while the primary sources of ALA come from plants such as nuts, seeds, and vegetable oils especially chia seeds, walnuts, flaxseed, canola, echium seed oils, and soybean oils (18, 31).

Bioavailability

There are different definitions for bioavailability. In a more specific context, bioavailability represents the rate and quantity at which a substance is absorbed by the intestinal tract and enters portal circulation (25). The bioavailability of omega-3 fatty acids depends on various factors such as chemical bonds, simultaneous food intake (especially its fat content), and the existence of additional elements affecting the uptake of omega-3 like calcium ions which reduce its availability (25). Appropriate functioning of pancreatic enzymes is the other important factor (32). The best way to assess short-term omega-3 availability is measuring its level in serum and plasma; while measuring the concentrations of EPA and DHA in red blood cell (RBC) membranes is a better way for long-term dietary intake and availability (33). Omega-3 can exist in free fatty acids (FFAs), phospholipid (PL), triacylglycerol (TAG), and ethyl ester (EE) forms. The FFA form exhibits greater bioavailability when compared to the EE form. On the other hand, ALA has low bioavailability because β-oxidation is the primary pathway for its utilization in the body and 85% of consumed ALA is lost due to the high rate of this process. In contrast, DHA has higher bioavailability due to less participation in β-oxidation. Also, fatty acids located at the sn-2 position of triacylglycerol have higher bioavailability due to easier absorption (34, 35).

Extraction

There are several techniques for extracting omega-3 oils from animal-based sources. One of them that was employed for the complete extraction of lipids from fish or other animal tissues is the chloroform-methanol extraction technique which requires specific ratios of chloroform ,methanol, and water as solvents (36, 37). Another technique used for omega-3 fatty acids-rich plant oil extraction is the cold-press technique. In this method, no heat is administered to the raw material prior to its passage through the conventional screw press, helping preserve higher levels of natural antioxidants that might be lost with other methods. For this reason, consumers believe that cold-pressed oils are natural and safe (38).

Supplement

Most omega-3 supplements are available over the counter, while some of them like, icosapent ethyl (Vascepa®) and EPA and DHA ethyl esters (Lovaza®), are prescription drugs (39). Different formulations of EPA and DHA are available in the form of a supplement including natural (typically derived from fish oil), reconstituted triglycerides (rTG), FFA (OM-3 FFA), and ethyl esters (OM-3 EE). These supplements are coated as capsules to be resistant to gastric acid and reduce side effects. They are usually in soft gel or TG oil forms as droplets trapped inside a gelatin matrix (gelled emulsions) formed by the microencapsulation method (40).

The maximum concentration of the supplement observed in plasma is defined as C_{max} (41). The determination of C_{max} for EPA and DHA can be accomplished between five to nine hours following oral administration (29). On the other hand, daily supplementation achieves stable levels of EPA and DHA in plasma within a two-week timeframe (42). The period required for EPA to reach its maximum absorption is approximately 2 weeks in plasma TGs, three weeks in serum cholesterol esters, around 2 months in RBCs, and over 12 months in most types of adipose tissue (43). DHA levels generally surpass EPA level in various body organs, such as the brain and retina (44). However, the incorporation of DHA into RBCs tends to occur at a slower rate compared to EPA (44). In addition, the EPA compound exhibits a half-life of 37 hours, while the DHA compound demonstrates a half-life of 48 hours (45).

Safety

According to government regulatory agencies, daily supplementation with omega-3 fatty acids up to a maximum of 3 to 4.5 grams per day is safe. In only 1 to 10% of individuals, it may cause side effects such as dyspepsia, nausea, diarrhea, and an increased tendency to bleed (46). The antithrombotic properties of omega-3 appear to upsurge the risk of bleeding in high doses (47, 48). So, it is recommended by the Food and Drug Administration (FDA) that the intake of EPA and DHA combination should not surpass 3 grams per day and a maximum of 2 grams should be from supplements (45). People who primarily obtain omega-3 from fish or are pregnant and breastfeeding should restrict their fish consumption to 2-4 servings per week because of their methyl mercury content and/or exchange fish varieties with elevated methylmercury levels,

such as swordfish, albacore tuna, dolphin fish, kingfish, and shark with fish with reduced levels of methylmercury, such as salmon, herring, sardines, and trout but DHA and EPA supplements do not have any methyl mercury (49).

Clinical Application: Anti-hyperlipidemia

Omega-3 fatty acids have a triglyceride-lowering effect (50). Omega-3 decreases lipid production by the liver and increases triglyceride clearance and β-oxidation (7, 51). Potential mechanisms of triglyceride-lowering properties of omega-3 fatty acids are related to the modulation of some transcription factors that are involved in lipid metabolism such as Sterol Regulatory Element-Binding Protein 1c (SREBP-1c) and Peroxisome Proliferator-Activated Receptor Gamma (PPAR-γ) (6). EPA suppresses SREBP-1c which plays a major role in the expression of genes that participate in fatty acid biosynthesis (7). Moreover, EPA activates PPAR-γ that enhances lipoprotein lipase (LPL) function (7).

Nervous System

Omega-3 fatty acids exhibit protective effects on the nervous system, that are vital for brain development and maintaining normal nervous system activity (52). Low intake of omega-3 is one of the risk factors for depression and omega-3 supplements may be helpful for the treatment of depression and schizophrenia (53, 54). Lower levels of omega-3 have been observed in individuals with autism spectrum disorder and Alzheimer's disease (53, 55). It has been suggested that the use of omega-3 supplements may have a beneficial impact on individuals with multiple sclerosis (MS) (56, 57). These fatty acids are correlated with the preservation of normal mental health and the low intake of them can lead to mental disorders due to their influence on nerve cell membrane fluidity or inflammatory processes (53).

Anti-inflammatory Effect

By increasing the intake of EPA and DHA intakes, their content in the phospholipids of cells that contribute to the process of inflammation increases (26). EPA and DHA intake decreases the production of prostaglandin E2 (PGE2), leukotriene B4, and thromboxane A2 by the inflammatory cells; thereby, they can modulate inflammation (58). In fact, a notable positive correlation exists between the ratio of arachidonic acid (ARA) to EPA in phospholipids of cells and PGE2 production (26). In fact, EPA and DHA intake results in a reduction in the production of pro-inflammatory cytokines such as Tumor Necrosis Factor-alpha (TNF-α), Interleukin-1β (IL-1β), and IL-6, while these fatty acids can increase the anti-inflammatory cytokines like IL-10 (59).

Omega-3 fatty acids likely exert a direct impact on inflammation by influencing the Nuclear Factor-kappa B (NF-κB) signaling pathway (26). Omega-3 decreases Lipopolysaccharide (LPS)-induced activation of NF-κB in monocytes, macrophages, and dendritic cells by reducing IκB phosphorylation (60, 61). In another pathway, omega-3 inhibits NF-κB by inducing PPAR-γ in dendritic cells, resulting in the diminished synthesis of pro-inflammatory cytokines TNF-α and IL-6 (26).

Anti-cancer Effect

Omega-3 exerts its anti-cancer effects in several ways. Omega-3 decreases angiogenesis by suppressing the Vascular Endothelial Growth Factor (VEGF) production, which increases vascular permeability, induces endothelial cell proliferation and migration, and stimulates endothelial cell survival (62). In another way, omega-3 inhibits inducible Nitric Oxide Synthase (iNOS), COX-2, and TNF-α gene expression as pro-angiogenic factors by blocking NF-κB and MAPK activation (62). In addition, it produces resolvins, protectins, and maresins as novel anti-inflammatory lipid mediators (63). Additional research is required to elucidate the mechanisms underlying the anti-cancer properties of omega-3 (63).

Autophagy: Definition

The term "autophagy" was coined by Belgian biochemist Christian de Duve, who was awarded the Nobel Prize in Physiology or Medicine in 1974 (64). Autophagy (a Greek term that means "eating of self") is a cellular mechanism that involves the breakdown and recycling of damaged organelles or other cellular components, including misfolded proteins (13, 65). It is crucial for survival and homeostasis (66). This mechanism is preserved throughout evolution, extending from yeast to humans (9). Presently, a set of autophagy-related genes (ATG) have been pinpointed through genetic screening in yeast (13). Remarkably, a substantial number of these genes are conserved not only in yeast but also in diverse organisms including plants, and mammals (13). Autophagy, through eliminating dysfunctional components, it plays a pivotal role in cellular quality control, prevention of neurodegeneration, adaptation to nutrient scarcity, tumor suppression, and removal of pathogens (65, 67). Furthermore, autophagy is involved in functions related to differentiation and development processes (65). The regulation of

autophagy is influenced by several factors including nutritional conditions, hormonal signals, and various cues like temperature, levels of oxygen, and cell density (66, 68).

Autophagy initiates with the creation and enlargement of an isolation membrane, also known as a phagophore. Consequently, the edges of the phagophore merge to create the autophagosome, a double-membraned vesicle that encloses cytoplasmic material (66). Autophagosome with various cargos ultimately merges with lysosomes that form autolysosomes. Within autolysosomes, the contents of the autophagosome undergo degradation facilitated by lysosomal hydrolases. Remained end products, including amino acids and other remnants from the degradation process, are transported back to the cytosol for potential reuse (9). Autophagy encompasses three primary types including macro-autophagy, micro-autophagy, and chaperone-mediated autophagy (CMA) (12). In macro-autophagy, portions of the cytoplasm or organelles are sequestered within autophagosomes and then degraded in the lysosomes (13). Micro-autophagy involves the direct engulfment of small portions of the cytoplasm by lysosomes (13). Finally, in chaperone-mediated autophagy (CMA), specific proteins are transported across the lysosomal membrane; while bound to chaperone proteins and proceed with the degradation (13).

The mTOR complexes serve as a vital point where various signaling pathways come together to control cellular homeostasis. AMP-activated protein kinase (AMPK) can inhibit the mTOR signaling pathway, which consequently activates unc-51 like autophagy activating kinase ½ (ULK1/2) complex which in turn activates autophagy. On the other hand, autophagy is suppressed by the PKB-AKT pathway that can activate mTOR. The class-III phosphatidylinositol 3-kinase (PI3K) complex controls the creation of the phagophore membrane. In addition, Beclin-1 plays a crucial role in membrane formation, and when it interacts with Bcl-2, suppresses the process of autophagy. Moreover, PI3P-binding domains contain specific proteins and their accumulation in the membrane leads to the incorporation of additional ATGs, which are essential for elongating and extending the autophagosome membrane. The two ubiquitin-like conjugation systems regulate the elongation process of the isolated membrane. In the first system, ATG7 activates ATG12, which facilitates the covalent attachment of ATG12 to the ATG5 protein. This coupling of ATG5 and ATG12 is followed by the recruitment of ATG16, resulting in the formation of the ATG12-ATG5-ATG16 complex. This assembled complex is subsequently recruited to the phagophore membrane. The second process involves the conjugation of the LC3 protein with a lipid molecule called phosphatidylethanolamine (PE). LC3 is initially synthesized as pro-LC3, and ATG4 cleaves it at the C-terminus to produce LC3-I. With the assistance of ATG7, ATG3 is attached to LC3-I to form LC3-II. Finally, the connection between LC3-II and PE facilitates the merging of autophagosomal membranes with lysosomes (9, 10, 67).

Autophagy and Some Diseases

Autophagy is associated with the pathogenesis of numerous diseases, including neurodegenerative disorders, cancer, and cardiovascular disease (66, 69). Certain genes are responsible for suppressing tumors and are situated upstream of the mTOR signaling pathway, which have the ability to activate autophagy. On the other hand, oncogene products trigger mTOR activation, which exhibits the ability to hinder autophagy; for instance, the frequently mutated p53 tumor suppressor gene might inhibit autophagy (70, 71), and the cellular proto-oncoproteins Bcl-2 and Bcl-XL, commonly elevated in human cancers, impede autophagy by binding to the autophagy protein Beclin 1 (72, 73). A robust association exists between molecules inducing autophagy and the prevention of tumors, as well as between molecules obstructing autophagy and the development of cancer (66). It appears that autophagy provides defense against various neurodegenerative disorders by removing misfolded proteins or eliminating aggregated mutant proteins linked to a range of distinct neurodegenerative conditions including Parkinson's disease and Alzheimer's disease (66, 74-76). Malfunctioning autophagy has been shown to be involved in inherited heart diseases like Danon and Pompe disease due to dysfunction in the autophagosome-lysosome fusion step. More importantly, autophagy might serve as a vital response to cardiac stresses such as ischemia or pressure overload, conditions commonly seen in patients with coronary artery disease, hypertension, aortic valvular disease, and congestive heart failure (66, 77).

Autophagy and Omega-3

Omega-3 can affect autophagy in several ways. It has been reported in a study on DU145 prostate cancer cell lines harboring mutant p53, ω3-PUFA DHA (50 μM DHA for 2 h) to be able to stimulate mitochondrial ROS generation and inhibit the Akt-mTOR signaling. Consequently, autophagy and apoptosis are induced and the p53-mutant cells' survival is decreased in prostate cancer (14) (Figure 1).
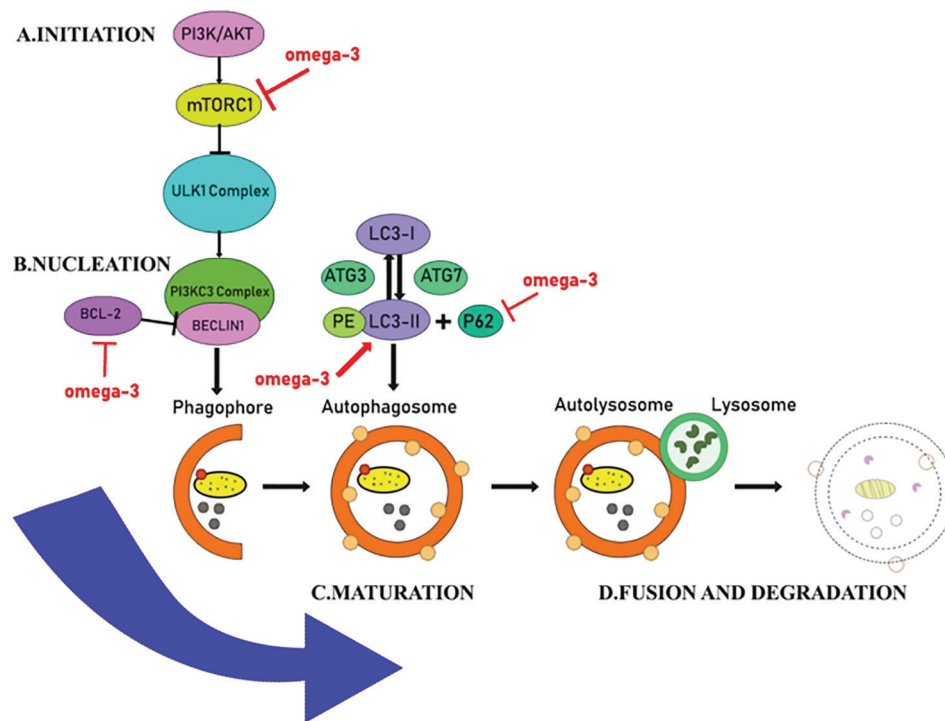
Figure 1: Effects of omega 3 supplementation on autophagy signaling pathways.

Similarly, pre-treatment with DHA (70 μM and 140 μM at 3 h) exhibited an augmentation in ROS and autophagy, thereby providing cellular rescue from cell cycle arrest provoked by misfolded proteins or oxidative stress in age-related macular degeneration (AMD) (78). It was shown that DHEA and EPEA which are derived from EPA and DHA enhanced PPARγ expression at a dose of 1 μM of DHEA or EPEA, as a result, AKT-mTOR pathways were inhibited. In addition, DHEA and EPEA treatment induced autophagy by persuading phosphorylation of Bcl-2 promoting its dissociation from beclin-1 in MCF-7 cell lines of breast cancer (79) (Figure 1).

Another study mentioned that autophagy regulated by mTOR necessitated intracellular calcium signaling. DHA (100 μM) increased the intracellular calcium by eliciting an intracellular calcium flux, so stimulated the formation of autophagosomes and enhanced autophagy (80). In addition, DHA treatment induced autophagy in glioblastoma multiform cells by increasing the LC3B-II level as a marker for autophagy in a dose-dependent manner (0 to 50 μM) (81) (Figure 1). In the same way, treatment with 100 μM DHA for 24 hours increased the LC3II formation and decreased p62 level, thus induced autophagy in Peripheral Blood Mononuclear Cells (PBMCs) and Dendritic Cells (DCs) in multiple myeloma (82) (Figure 1).

It was shown that the administration of LPS could consistently hinder the contractile function of the left ventricle and induced cardiac injury in wild-type mice. However, this effect was not significantly observed in fat-1 mice, which possessed an increased concentration of endogenous ω-3 PUFAs. The fat-1 mice exhibited notably higher level of LC3 II / I, as well as increased expression of PPAR-γ and sirtuin (Sirt)-1; while concurrently experiencing a reduction in NFκB activation. These alterations resulted in an augmentation of autophagic flux activities (83). Lipophagy, a type of selective autophagy was demonstrated to play a pivotal role in the regulation of lipid reserves in the liver and adipose tissue (84). When L02 hepatocytes were treated with 1 mM FFA, cellular lipid percentage and apoptotic cells increased when compared to the control group. Suppressing autophagy resulted in an excessive accumulation of lipids in response to FFA induction. However, omega-3 fatty acids (0.1 mM EPA or DHA) decreased the percentage of cells that underwent apoptosis and induced autophagy by down-regulating the expression of Stearoyl-CoA Desaturase1 in hepatocytes and increase in LC3II/LC3I, thereby exerting a protective effect on these cells in Non-alcoholic Fatty Liver Disease (NAFLD) (85).

The possible mechanisms also could be modulating gene transcription of lipid metabolism-related genes, including peroxisome proliferator-activated receptor alpha (PPAR-α), Liver X Receptor (LXR), hepatic nuclear factor-4a, Carbohydrate Responsive Element Binding Protein (ChREBP), and SREBP 1c, and reduction in the expression of pro-inflammatory molecules, namely TNF-α and IL-6, as well as ROS by Omega 3 fatty acids (85-87). A recent investigation demonstrated that TNF-α, a

pro-inflammatory cytokine implicated in obesity to be able to diminish the expression of p62 protein and elevate the LC3II/LC3I ratio, thereby inducing autophagy (Figure 1). Conversely, Maresin 1 (100 nM), a lipid mediator originating from omega-3, was found to decrease the LC3II/LC3I ratio, a crucial indicator of autophagosome formation, thereby impeding autophagy (88).

In the context of pancreatic cancer, it has been observed that EPA (10 mM) triggers autophagy through the accumulation of ROS, while concurrently reducing its capacity to induce apoptotic cell death. Consequently, it has been proposed that the combination of EPA with an autophagy inhibitor could potentially serve as a valuable approach to enhance the therapeutic efficacy in the treatment of pancreatic cancer (7). Omega-3 PUFAs can enhance autophagy and protect against renal injury. In a unilateral ureteral obstruction (UUO) model of chronic kidney disease (CKD), DHA and EPA treatment of human proximal tubular cells (HK2) increased autophagic flux and AMPK phosphorylation and reduced epithelial-mesenchymal transition. In vivo, ω3-PUFAs reduced oxidative stress, inflammation, and fibrosis in UUO mice, while promoting autophagic markers (LC3-II, Beclin-1, Atg7) and reducing p62. These results suggested ω3-PUFAs to activate autophagy and offer protection against kidney injury (89).

On the other hand, it was shown that EPA (60 μg/mL) or DHA (50 μg/ml) treatment for 24 h decreased Beclin-1 expression levels and inhibited autophagosome formation in A549 lung cancer cells. Also, these PUFAs increased p-Akt and p-mTOR levels, resulting in activation of Akt-mTOR pathways and a reduction in autophagy during the early stage. In addition, this treatment for 24 h increased p62 level and finally a blockage in the autophagic flux. Therefore, EPA and DHA exerted anti-proliferative effects in A549 lung cancer cells by inhibiting of autophagy and induction of apoptosis (15). Also, colistin (a potent antibiotic)-induced nephrotoxicity limited its clinical use. A study revealed the protective effect of omega-3 nanoemulsion in rats, with a focus on autophagy. Colistin caused renal dysfunction, oxidative stress, inflammation, and histological damage, along with increased autophagy markers like LC3-II, Beclin-1 and decreased p62. Omega-3 nanoemulsion reversed these effects by modulating autophagy, reducing oxidative stress, and improving renal function. Therefore, omega-3 nanoemulsion could protect against colistin-induced nephrotoxicity, primarily through the regulation of autophagy and related pathways (90).

Conclusion
Omega-3 fatty acids play a vital role in modulating autophagy, offering potential benefits as an adjunct therapy in combating diseases. They induce autophagy through ROS generation, suppression of the mTOR signaling pathway, and the modulation of autophagy-related proteins. These fatty acids exhibit anti-inflammatory, anti-cancer, and neuroprotective properties, making them valuable for health. However, these effects can be different in various doses and settings. Also, their effects on autophagy can vary depending on the specific conditions, requiring further research to understand the exact underlying mechanisms. Omega-3 fatty acids and autophagy interaction was demonstrated to hold promise for innovative therapeutic strategies in disease management and overall well-being. More experimental and clinical trial studies are needed to substantiate their clinical application.

Authors' Contribution
SEK: Hypothesized the review. RT and MZ: Contributed to the systematic search and data extraction. RT, MZ and AZ: Contribute to manuscript drafting. SEK: Supervised the study. All authors approved the final manuscript for submission.

Conflict of Interest
The authors confirm no conflicts of interest.

References
1   Mehrabani D, Masoumi SJ, Masoumi AS, et al. Role of Diet in Mesenchymal Stem Cells' Function: A Review. *Int J Nutr Sci*. 2023;8:9-19. DOI: 10.30476/IJNS.2023.97788.1221.

2   Zailani H, Satyanarayanan SK, Liao WC, et al. Omega-3 Polyunsaturated Fatty Acids in Managing Comorbid Mood Disorders in Chronic Obstructive Pulmonary Disease (COPD): A Review. *J Clin Med*. 2023;12:2653. DOI: 10.3390/jcm12072653. PMID: 37048736

3   Zhu S, Lin G, Song C, Wu Y, Feng N, Chen W, et al. RA and ω-3 PUFA co-treatment activates autophagy in cancer cells. *Oncotarget*. 2017;8:109135. DOI: 10.18632/oncotarget.22629.

PMID: 29312596.

4 Gammone MA, Riccioni G, Parrinello G, et al. Omega-3 polyunsaturated fatty acids: Benefits and endpoints in sport. *Nutrients*. 2019;11:46. DOI: 10.3390/nu11010046. PMID: 30591639.

5 Chalifoux NV, Silverstein DC, Page B, Fudge JMJAJoVR. Assessment of changes in intra-abdominal pressure and abdominal perfusion pressure in late-term pregnant queens undergoing elective ovariohysterectomy. *Am J Vet Res*. 2022;83: ajvr.22.02.0023. DOI: 10.2460/ajvr.22.02.0023. PMID: 35895779.

6 Yazdani J, Dehzad MJ, Ommati MM, et al. The Effect of Banana (Musa nana Lour.) Peel Extract and Omega-3 on Biochemical and Histopathological Characteritics in Rat Model of Polycystic Ovary Syndrome. *Int J Nutr Sci*. 2025;10:93-103. DOI: 10.30476/ijns.2025.101331.1297.

7 Shibabaw TJM, Biochemistry C. Omega-3 polyunsaturated fatty acids: Anti-inflammatory and anti-hypertriglyceridemia mechanisms in cardiovascular disease. *Mol Cell Biochem*. 2021;476:993-1003. DOI: 10.1007/s11010-020-03965-7. PMID: 33179122.

8 Hassanshahi N, Masoumi SJ. The Effect of Omega-3 Fatty Acids in Ulcerative Colitis: A Systematic Review. *Int J Nutr Sci*. 2018;3:58-64.

9 Kocaturk NM, Akkoc Y, Kig C, et al. Autophagy as a molecular target for cancer treatment. *Eur J Pharm Sci*. 2019;134:116-37. DOI: 10.1016/j.ejps.2019.04.011. PMID: 30981885.

10 Abroudi M, Mehrabani D, Zare S, et al. In Vitro Assessment of Morphology, Proliferation, Apoptosis and Differential Potential of Dental Pulp Stem Cells, When Marijuana Is Added to Nutrients of Cell Culture Medium. *Int J Nutr Sci*. 2024;9:62-70. DOI: 10.30476/IJNS.2024.101034.1288.

11 Levine B, Kroemer G. Biological functions of autophagy genes: a disease perspective. *Cell*. 2019;176:11-42. DOI: 10.1016/j.cell.2018.09.048. PMID: 30633901.

12 Levy JMM, Towers CG, Thorburn A. Targeting autophagy in cancer. *Nat Rev Cancer*. 2017;17:528-42. DOI: 10.1038/nrc.2017.53. PMID: 28751651.

13 Glick D, Barth S, Macleod KF. Autophagy: cellular and molecular mechanisms. *J Pathol*. 2010;221:3-12. DOI: 10.1002/path.2697. PMID: 20225336.

14 Shin S, Jing K, Jeong S, et al. The omega-3 polyunsaturated fatty acid DHA induces simultaneous apoptosis and autophagy via mitochondrial ROS-mediated Akt-mTOR signaling in prostate cancer cells expressing mutant p53. *BioMed Res Int*. 2013;2013:568671. DOI: 10.1155/2013/568671. PMID: 23841076.

15 Yao Q, Fu T, Wang L, et al. Role of autophagy in the ω-3 long chain polyunsaturated fatty acid-induced death of lung cancer A549 cells. *Oncol Lett*. 2015;9:2736-42. DOI: 10.3892/ol.2015.3110. PMID: 26137138.

16 Anari L, Mehrabani D, Nasiri M, et al. in Vitro Effect of Methamphetamine on Proliferation, Differentiation and Apoptosis of Adipose Tissue Stem Cells. *J Pharm Pharm Sci*. 2022;25:69-76. DOI: 10.18433/jpps31843. PMID: 35030074.

17 Andisheh Tadbir A, Mehrabani D, Heydari ST. Primary malignant tumors of orofacial origin in Iran. *J Craniofac Surg*. 2008;19:1538-41. DOI: 10.1097/SCS.0b013e31818eeccb. PMID: 19098546.

18 Shahidi F, Ambigaipalan P. Omega-3 polyunsaturated fatty acids and their health benefits. *Annu Rev Food Sci Technol*. 2018;9:345-81. DOI: 10.1146/annurev-food-111317-095850. PMID: 29350557.

19 Saenz de Viteri M, Hernandez M, Bilbao-Malavé V, et al. A Higher proportion of eicosapentaenoic acid (EPA) when combined with docosahexaenoic acid (DHA) in omega-3 dietary supplements provides higher antioxidant effects in human retinal cells. *Antioxidants (Basel)*. 2020;9:828. DOI: 10.3390/antiox9090828. PMID: 32899655.

20 Babajafari S, Moosavi-Nasab M, Nasrpour S, et al. Comparison of Enzymatic Hydrolysis and Chemical Methods for Oil Extraction from Rainbow Trout (Oncorhynchus Mykiss) Waste and Its Influence on Omega 3 Fatty Acid Profile. *Int J Nutr Sci*. 2017;2:58-65.

21 Deckelbaum RJ, Torrejon CJTJon. The omega-3 fatty acid nutritional landscape: health benefits and sources. *J Nutr*. 2012;142:587S-91S. DOI: 10.3945/jn.111.148080. PMID: 22323763.

22 Aghasadeghi K, Zarei-Nezhad M, Keshavarzi A, et al. The prevalence of coronary risk factors in Iranian lor migrating tribe. *Arch Iran Med*. 2008;11:322-5. PMID: 18426325.

23 Buckley JD, Howe PRJN. Long-chain omega-3 polyunsaturated fatty acids may be beneficial for reducing obesity—a review. *Nutrients*. 2010;2:1212-30. DOI: 10.3390/nu2121212. PMID: 22254005.

24 Chauhan S, Kodali H, Noor J, et al. Role of Omega-3 Fatty Acids on Lipid Profile in Diabetic Dyslipidaemia: Single Blind, Randomised Clinical Trial. *J Clin Diagn Res*. 2017;11:Oc13-oc6. DOI: 10.7860/jcdr/2017/20628.9449. PMID: 28511427.

25 Punia S, Sandhu KS, Siroha AK, et al. Omega

3-metabolism, absorption, bioavailability and health benefits–A review. *Pharma Nutr.* 2019;10:100162. DOI: 10.1016/j.phanu.2019.100162.

26 Calder PC. Omega-3 fatty acids and inflammatory processes: from molecules to man. *Biochem Soc Trans.* 2017;45:1105-15. DOI: 10.1042/BST20160474. PMID: 28900017.

27 Zhang X, Yang N, Ai D, et al. Systematic metabolomic analysis of eicosanoids after omega-3 polyunsaturated fatty acid supplementation by a highly specific liquid chromatography–tandem mass spectrometry-based method. *J Proteome Res.* 2015;14:1843-53. DOI: 10.1021/pr501200u. PMID: 25736083.

28 Murru E, Banni S, Carta G. Nutritional properties of dietary omega-3-enriched phospholipids. *Biomed Res Int.* 2013;2013:965417. DOI: 10.1155/2013/965417. PMID: 23984423.

29 Cholewski M, Tomczykowa M, Tomczyk M. A comprehensive review of chemistry, sources and bioavailability of omega-3 fatty acids. *Nutrients.* 2018;10:1662. DOI: 10.3390/nu10111662. PMID: 30400360.

30 Fu Y, Wang Y, Gao H, et al. Associations among dietary omega-3 polyunsaturated fatty acids, the gut microbiota, and intestinal immunity. *Mediators Inflamm.* 2021;2021:8879227. DOI: 10.1155/2021/8879227. PMID: 33488295.

31 DeFilippis AP, Sperling LS. Understanding omega-3's. *Am Heart J.* 2006;151:564-70. DOI: 10.1016/j.ahj.2005.03.051. PMID: 16504616.

32 Blair HA, Dhillon S. Omega-3 carboxylic acids (Epanova): a review of its use in patients with severe hypertriglyceridemia. *Am J Cardiovasc Drugs.* 2014;14:393-400. DOI: 10.1007/s40256-014-0090-3. PMID: 25234378.

33 Schuchardt JP, Hahn A. Bioavailability of long-chain omega-3 fatty acids. *Prostaglandins Leukot Essent Fatty Acids.* 2013;89:1-8. DOI: 10.1016/j.plefa.2013.03.010. DOI: 10.1016/j.plefa.2013.03.010. PMID: 23676322.

34 Libinaki R, Gavin PD. Changes in bioavailability of omega-3 (DHA) through Alpha-Tocopheryl Phosphate Mixture (TPM) after oral administration in rats. *Nutrients.* 2017;9:1042. DOI: 10.3390/nu9091042. PMID: 28930161.

35 Tengku-Rozaina TM, Birch EJ. Enrichment of omega-3 fatty acids of refined hoki oil. *J Am Oil Chem Society.* 2013;90:1111-9. DOI: 10.1007/s11746-013-2260-x.

36 Bennett PM, Weber LP, Janz DM. Comparison of chloroform–methanol-extracted and solvent-free triglyceride determinations in four fish species. *J Aquat Anim Health.* 2007;19:179-85. DOI:

10.1577/H06-044.1. PMID: 18201059.

37 Deepika D, Vegneshwaran V, Julia P, et al. Investigation on oil extraction methods and its influence on omega-3 content from cultured salmon. *J Food Process Technol.* 2014;5:1-13. DOI: 10.4172/2157-7110.1000401.

38 Parker TD, Adams D, Zhou K, et al. Fatty acid composition and oxidative stability of cold-pressed edible seed oils. *J Food Sci.* 2003;68:1240-3. DOI:10.1111/j.1365-2621.2003.tb09632.x.

39 Mohebi-Nejad A, Bikdeli B. Omega-3 supplements and cardiovascular diseases. *Tanaffos.* 2014;13:6-14. PMID: 25191488.

40 Tamargo J, Tamargo M. Pharmacokinetics and safety profile of omega-3 polyunsaturated fatty acids. Omega-3 Fatty Acids: Keys to Nutritional Health. 2016.p.541-84. DOI:10.1007/978-3-319-40458-5_39.

41 Qin Y, Nyheim H, Haram EM, et al. A novel self-micro-emulsifying delivery system (SMEDS) formulation significantly improves the fasting absorption of EPA and DHA from a single dose of an omega-3 ethyl ester concentrate. *Lipids Health Dis.* 2017;16:204. DOI: 10.1186/s12944-017-0589-0. PMID: 29037249.

42 Cicero AF, Colletti A. Krill oil: evidence of a new source of polyunsaturated fatty acids with high bioavailability. Taylor & Francis; 2015.p.1-4.

43 Walker CG, Browning LM, Mander AP, et al. Age and sex differences in the incorporation of EPA and DHA into plasma fractions, cells and adipose tissue in humans. *Br J Nutr.* 2014;111:679-89. DOI: 10.1017/S0007114513002985. PMID: 24063767.

44 Fabian CJ, Kimler BF, Hursting SD. Omega-3 fatty acids for breast cancer prevention and survivorship. *Breast Cancer Res.* 2015;17:62. DOI: 10.1186/s13058-015-0571-6. PMID: 25936773.

45 Krupa K, Fritz K, Parmar M. Omega-3 fatty acids. 2020.

46 Politano CA, López-Berroa J. Omega-3 Fatty Acids and Fecundation, Pregnancy and Breastfeeding. *Rev Bras Ginecol Obstet.* 2020;42:160-4. DOI: 10.1055/s-0040-1708090. PMID: 32232824.

47 Bays HE. Safety considerations with omega-3 fatty acid therapy. *Am J Cardiol.* 2007;99:35c-43c. DOI: 10.1016/j.amjcard.2006.11.020. PMID: 17368277.

48 Krause MV, Mahan LK. Krause and Mahan's Food & the Nutrition Care Process: Elsevier; 2021.

49 Mozaffarian D, Rimm EB. Fish intake,

contaminants, and human health: evaluating the risks and the benefits. JAMA. 2006;296:1885-99. DOI: 10.1001/jama.296.15.1885. PMID: 17047219.

50 Luna-Castillo KP, Olivares-Ochoa XC, et al. The effect of dietary interventions on hypertriglyceridemia: From public health to molecular nutrition evidence. *Nutrients*. 2022;14:1104. DOI: 10.3390/nu14051104. PMID: 35268076.

51 Harris WS, Miller M, Tighe AP, et al. Omega-3 fatty acids and coronary heart disease risk: clinical and mechanistic perspectives. *Atherosclerosis*. 2008;197:12-24. DOI: 10.1016/j. atherosclerosis.2007.11.008. PMID: 18160071.

52 Mazza M, Pomponi M, Janiri L, et al. Omega-3 fatty acids and antioxidants in neurological and psychiatric diseases: an overview. *Prog Neuropsychopharmacol Biol Psychiatry*. 2007;31:12-26. DOI: 10.1016/j.pnpbp.2006.07.010. PMID: 16938373.

53 Lange KW. Omega-3 fatty acids and mental health. *Global Health J*. 2020;4:18-30. DOI: 10.1016/j.glohj.2020.01.004.

54 Pawełczyk T, Grancow M, Kotlicka-Antczak M, et al. Omega-3 fatty acids in first-episode schizophrenia - a randomized controlled study of efficacy and relapse prevention (OFFER): rationale, design, and methods. *BMC Psychiatry*. 2015;15:97. DOI: 10.1186/s12888-015-0473-2. PMID: 25934131.

55 Malekzadeh S, Edalatmanesh MA, Mehrabani D, et al. Dental Pulp Stem Cells Transplantation Improves Passive Avoidance Memory and Neuroinflammation in Trimethyltin-Induced Alzheimer's Disease Rat Model. *Galen Med J*. 2021;10:e2254. DOI:10.31661/gmj.v10i0.2254.

56 AlAmmar WA, Albeesh FH, Ibrahim LM, et al. Effect of omega-3 fatty acids and fish oil supplementation on multiple sclerosis: a systematic review. *Nutr Neurosci*. 2021;24:569-79. DOI: 10.1080/1028415x.2019.1659560. PMID: 31462182.

57 Mehrabani G, Aminian S, Mehrabani G, et al. Dietetic Plans within the Multiple Sclerosis Community: A Review. *Int J Nutr Sci*. 2019;4:14-22. DOI: 10.30476/IJNS.2019.81531.1007.

58 Simopoulos AP. Omega-3 fatty acids in inflammation and autoimmune diseases. J *Am Coll Nutr*. 2002;21:495-505. DOI: 10.1080/07315724.2002.10719248. PMID: 12480795.

59 Calder PC. Marine omega-3 fatty acids and inflammatory processes: Effects, mechanisms and clinical relevance. *Biochim Biophys Acta*. 2015;1851:469-84. DOI: 10.1016/j. bbalip.2014.08.010. PMID: 25149823.

60 Weatherill AR, Lee JY, Zhao L, et al. Saturated and polyunsaturated fatty acids reciprocally modulate dendritic cell functions mediated through TLR4. *J Immunol*. 2005;174:5390-7. DOI: 10.4049/jimmunol.174.9.5390. PMID:15843537.

61 Lee JY, Sohn KH, Rhee SH, et al. Saturated fatty acids, but not unsaturated fatty acids, induce the expression of cyclooxygenase-2 mediated through Toll-like receptor 4. *J Biol Chem*. 2001;276:16683-9. DOI: 10.1074/jbc. M011695200. PMID: 11278967.

62 Spencer L, Mann C, Metcalfe M, Webb M, Pollard C, Spencer D, et al. The effect of omega-3 FAs on tumour angiogenesis and their therapeutic potential. Eur J Cancer. 2009;45(12):2077-86. Epub 20090601. doi: 10.1016/j.ejca.2009.04.026. PubMed PMID: 19493674.

63 Hull MA. Omega-3 polyunsaturated fatty acids. *Best Pract Res Clin Gastroenterol*. 2011;25:547-54. DOI: 10.1016/j.bpg.2011.08.001. PMID: 22122770.

64 Li X, He S, Ma B. Autophagy and autophagy-related proteins in cancer. *Mol Cancer*. 2020;19:12. DOI: 10.1186/s12943-020-1138-4. PMID: 31969156.

65 Mizushima N, Levine B. Autophagy in mammalian development and differentiation. *Nat Cell Biol*. 2010;12:823-30. DOI: 10.1038/ncb0910-823. PMID: 20811354.

66 Levine B, Kroemer G. Autophagy in the pathogenesis of disease. *Cell*. 2008;132:27-42. DOI: 10.1016/j.cell.2007.12.018. PMID: 18191218.

67 Mizushima N, Yoshimori T, Levine B. Methods in mammalian autophagy research. *Cell*. 2010;140:313-26. DOI: 10.1016/j.cell.2010.01.028. PMID: 20144757.

68 Zare M, Naeimi S, Zekri S, et al. What Happens When Methamphetamine Is Added to Nutrients of Culture Medium: In Vitro Effects on Proliferation, Differentiation and Apoptosis of Human Fibroblast Cells. *Int J Nutr Sci*. 2024;9:194-202. DOI: 10.30476/ijns.2024.102840.1328.

69 Mohammadzadeh N, Mehrabani D, Zare S, et al. What Happens When Methamphetamine Is Added to Nutrients of Cell Culture Medium? In Vitro Assessment of Morphological, Growth and Differential Potential of Wharton's Jelly Stem Cells. *Int J Nutr Sci*. 2022;7:233-240. DOI: 10.30476/ijns.2022.97432.1210.

70 Crighton D, Wilkinson S, O'Prey J, et al. DRAM, a p53-induced modulator of autophagy, is critical for apoptosis. *Cell*. 2006;126:121-34. DOI: 10.1016/j.cell.2006.05.034. PMID: 16839881.

71 Feng Z, Zhang H, Levine AJ, et al. The coordinate regulation of the p53 and mTOR pathways in cells. *Proc Natl Acad Sci U S A.* 2005;102:8204-9. DOI: 10.1073/pnas.0502857102. PMID: 15928081.

72 Pattingre S, Tassa A, Qu X, et al. Bcl-2 antiapoptotic proteins inhibit Beclin 1-dependent autophagy. *Cell.* 2005;122:927-39. DOI: 10.1016/j.cell.2005.07.002. PMID: 16179260.

73 Maiuri MC, Le Toumelin G, Criollo A, et al. Functional and physical interaction between Bcl-XL and a BH3-like domain in Beclin-1. *EMBO J.* 2007;26:2527-39. DOI: 10.1038/sj.emboj.7601689. PMID: 17446862.

74 Williams A, Jahreiss L, Sarkar S, et al. Aggregate-prone proteins are cleared from the cytosol by autophagy: therapeutic implications. *Curr Top Dev Biol.* 2006;76:89-101. DOI: 10.1016/S0070-2153(06)76003-3. PMID: 17118264.

75 Rubinsztein DC, Gestwicki JE, Murphy LO, et al. Potential therapeutic applications of autophagy. *Nat Rev Drug Discov.* 2007;6:304-12. DOI: 10.1038/nrd2272. PMID: 17396135.

76 Malekzadeh S, Edalatmanesh MA, Mehrabani D, et al. Drugs induced alzheimer's disease in animal model. *Galen Med J.* 2017;6:185-96. DOI: 10.22086/gmj.v6i3.820.

77 Terman A, Brunk UT. Autophagy in cardiac myocyte homeostasis, aging, and pathology. *Cardiovasc Res.* 2005;68:355-65. DOI: 10.1016/j.cardiores.2005.08.014. PMID: 16213475.

78 Johansson I, Monsen VT, Pettersen K, et al. The marine n-3 PUFA DHA evokes cytoprotection against oxidative stress and protein misfolding by inducing autophagy and NFE2L2 in human retinal pigment epithelial cells. *Autophagy.* 2015;11:1636-51. DOI: 10.1080/15548627.2015.1061170. PMID: 26237736.

79 Rovito D, Giordano C, Vizza D, et al. Omega-3 PUFA ethanolamides DHEA and EPEA induce autophagy through PPARγ activation in MCF-7 breast cancer cells. *J Cell Physiol.* 2013;228:1314-22. DOI: 10.1002/jcp.24288. PMID: 23168911.

80 Williams-Bey Y, Boularan C, Vural A, et al. Omega-3 free fatty acids suppress macrophage inflammasome activation by inhibiting NF-κB activation and enhancing autophagy. *PLoS One.* 2014;9:e97957. DOI: 10.1371/journal.pone.0097957. PMID: 24911523.

81 Kim S, Jing K, Shin S, et al. ω3-polyunsaturated fatty acids induce cell death through apoptosis and autophagy in glioblastoma cells: In vitro and in vivo. *Oncol Rep.* 2018;39:239-46. DOI: 10.3892/or.2017.6101. PMID: 29192322.

82 D'Eliseo D, Di Renzo L, Santoni A, et al. Docosahexaenoic acid (DHA) promotes immunogenic apoptosis in human multiple myeloma cells, induces autophagy and inhibits STAT3 in both tumor and dendritic cells. *Genes Cancer.* 2017;8:426. DOI: 10.18632/genesandcancer.131.PMID: 28435516.

83 Mao S, Ma H, Chen P, et al. Fat-1 transgenic mice rich in endogenous omega-3 fatty acids are protected from lipopolysaccharide-induced cardiac dysfunction. *ESC Heart Failure.* 2021;8:1966-78. DOI: 10.1002/ehf2.13262. PMID: 33665922.

84 Christian P, Sacco J, Adeli K. Autophagy: Emerging roles in lipid homeostasis and metabolic control. *Biochim Biophys Acta.* 2013;1831:819-24. DOI: 10.1016/j.bbalip.2012.12.009. PMID: 23274236.

85 Chen Y, Xu C, Yan T, et al. ω-3 Fatty acids reverse lipotoxity through induction of autophagy in nonalcoholic fatty liver disease. *Nutrition.* 2015;31:1423-9. DOI: 10.1016/j.nut.2015.05.022. PMID: 26429665.

86 Pettinelli P, Obregón A, Videla L. Molecular mechanisms of steatosis in nonalcoholic fatty liver disease. *Nutr Hosp.* 2011;26:441-50. DOI: 10.1590/S0212-16112011000300003. PMID: 21892559.

87 Yeon JE, Choi KM, Baik SH, et al. Reduced expression of peroxisome proliferator-activated receptor-α may have an important role in the development of non-alcoholic fatty liver disease. *J Gastroenterol Hepatol.* 2004;19:799-804. DOI: 10.1111/j.1440-1746.2004.03349.x. PMID: 15209628.

88 Laiglesia LM, Lorente-Cebrián S, López-Yoldi M, et al. Maresin 1 inhibits TNF-alpha-induced lipolysis and autophagy in 3T3-L1 adipocytes. *J Cell Physiol.* 2018;233:2238-46. DOI: 10.1002/jcp.26096. PMID: 28703289.

89 Han S, Choi H, Park H, et al. Omega-3 Fatty Acids Attenuate Renal Fibrosis via AMPK-Mediated Autophagy Flux Activation. *Biomedicines.* 2023;11:2553. DOI: 10.3390/biomedicines11092553. PMID: 37760994.

90 Taha M, Abdelbagi O, Baokbah TAS, et al. Insights into the protective effect of omega-3 nanoemulsion against colistin-induced nephrotoxicity in experimental rats: regulation of autophagy and necroptosis via AMPK/mTOR and RIPK1/RIPK3/MLKL signaling pathways. *Ren Fail.* 2024;46:2429686. DOI: 10.1080/0886022x.2024.2429686. PMID: 39584420.