Omega-3 Fatty Acids in Inflammatory Diseases

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Introduction

Fatty acids are straight chain hydrocarbons with a carboxyl group at one end and a methyl group at the other. The carboxylic acid (-COOH) end is considered as the beginning of the chain, thus designated as "alpha," and the methyl ($-CH_3$) end is considered the "tail" of the chain, designated as "omega." There are three major classes of fatty acids, namely saturated fatty acids, monounsaturated fatty acids (MUFA), and polyunsaturated fatty acids (PUFAs). The saturated fatty acids do not contain any double bonds within the acyl chain, while unsaturated fatty acids contain at least one double bond. When a single double bond is present within the acyl chain, it is called as MUFA, and when two or more double bonds are present, they are referred to as PUFAs. The PUFAs can be further classified as omega-3 fatty acids (also called ω-3 fatty acids or *n*-3 fatty acids) and omega-6 fatty acids (also called ω-6 fatty acids or *n*-6 fatty acids) based on the location of the first double bond from the terminal methyl end of the molecule. Omega-3 fatty acids possess first double bond (C = C) at the third carbon atom from the methyl end of the carbon chain, while omega-6 fatty acids have first double bond (C = C) at the sixth carbon atom from the methyl end of the carbon chain. The human body can produce all except two of the fatty acids it requires, i.e., linoleic acid and α-linolenic acid, as the enzymes (desaturases) required to introduce double bonds in the n-3 and n-6 positions are not present in mammals. Linoleic acid (LA, C18:2n-6) is the precursor to the n-6 series of fatty acids and α-linolenic acid (ALA, C18:3n-3) is the precursor to the n-3 series of fatty acids. These fatty acids must therefore be obtained from the diet, and accordingly, they are known as essential fatty acids.

The essential fatty acids from omega-3 series involved in human physiology are alpha-linolenic acid (ALA), eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHA). Marine algae and phytoplankton are primary sources of omega-3 fatty acids. Common sources of plant oils containing ALA fatty acid include walnut, edible seeds, clary sage seed oil, algal oil, flaxseed oil, Sacha Inchi oil, *Echium* oil, and hemp oil, while sources of animal EPA and DHA fatty acids are mostly found in seafood, but fish do not actually produce these fatty acids. In fact, these compounds are produced by single-cell marine organisms that are consumed by fish. Other sources include egg oil, squid oils, and krill oil.

PUFAs regulate a wide variety of biological functions, depending on the location of the last double bond, which range from blood pressure and blood clotting to the correct development and functioning of the brain and nervous system [1]. In addition, lipid mediators generated from long-chain (LC-) PUFA (arachidonic acid (AA) in the n-6 series and EPA and DHA in the n-3 series) have important roles in immune regulation and inflammation [2]. This chapter is focused to give an insight about how dietary omega-3 fatty acids help in the management of inflammatory disorders.

Immune System and Inflammation

The immune system provides protection from an array of infectious agents while permitting tolerance to self-antigens and non-threatening agents such as food proteins and bacterial gut flora. The body's immediate response to infection or injury begins with inflammation. Inflammation functions to begin the process of elimination of invading pathogens and toxins and to repair damaged tissue. The immune

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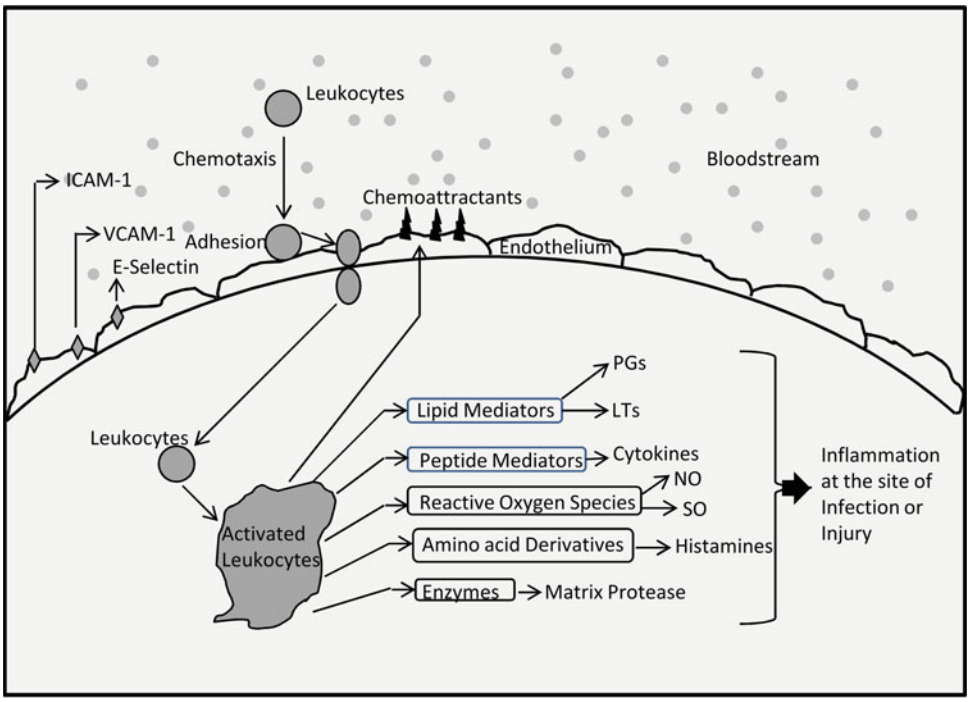

Fig. 11.1 Diagrammatic representation of the immunologic responses to infection and injury (Modified from [4]) © American Society for Nutrition. *PGs* Prostaglandins, *LTs* Leukotrienes, *NO* Nitric oxide, *SO* Superoxide, *ICAM-1* Intercellular adhesion molecule, *VCAM-1* Vascular Cell adhesion molecule

response involves a complexity of blood-borne factors and different immune cells with different roles but they act together to create a highly regulated and well coordinated immune response [3]. Clinical characteristics of acute inflammation include redness, swelling, heat, and pain. These occur as a result of increased blood flow to the site of inflammation; increased permeability across blood capillaries caused by retraction of endothelial cells, which allows large molecules (e.g., complement, antibodies, and cytokines) to leave the bloodstream and cross the endothelial wall; increased movement of leukocytes from the bloodstream into the surrounding tissue and then to the site of inflammation as depicted in Fig. 11.1. This movement is induced by release of chemoattractants and by the upregulation of adhesion molecules such as intercellular adhesion molecule-1 (ICAM-1), vascular cell adhesion molecule-1 (VCAM-1), and E-selectin on the surface of endothelial cells allowing leukocyte binding and subsequent diapedesis. The last stage involves release of mediators from leukocytes at the site of inflammation. These may include lipid mediators (e.g., prostaglandins [PGs], leukotrienes [LTs]), peptide mediators (e.g., cytokines), reactive oxygen species (e.g., superoxide), amino acid derivatives (e.g., histamine), and enzymes (e.g., matrix proteases) depending upon the cell type involved, the nature of the inflammatory stimulus, the anatomical site involved, and the stage during the inflammatory response. Several of these mediators may act to amplify the inflammatory process by acting as chemoattractants. Some of the inflammatory mediators may escape the inflammatory site into the circulation and from there they can exert systemic effects. Thus, inflammation and the inflammatory response are part of the normal, innate immune response [4].

Although inflammation is a normal response, when it occurs in an uncontrolled or inappropriate manner excessive damage to host tissues and disease can ensue. Such uncontrolled or inappropriate inflammatory responses are characterized by hyperexpression of endothelial and leukocyte adhesion molecules, appearance of soluble forms of adhesion molecules in the circulation, sequestration of leukocytes to sites where they are not usually found, production of inflammatory mediators, and damage to host tissues [4].

Role of PUFAS in Inflammation

The major substrates for energy production are fatty acids; however, they are also involved in the formation of cellular structures as well as in the transmission of cellular signals. Dietary lipids are absorbed and distributed to essentially every cell membrane in the body where they perform important structural and functional roles. They are known to modulate the immune system by various means, such as altering membrane fluidity, regulating eicosanoid metabolites, oxidative stress, producing lipid peroxides, regulation of gene expression, apoptosis or modulation of gastrointestinal microbiota, and interacting directly with cellular activation processes [3]. Polyunsaturated fatty acids and

their metabolites are crucial to the physiologic and pathophysiologic processes in inflammation. Altering fatty acid type and their composition in phospholipids of immune cells through diet supplements for beneficial outcomes in disease has been of major interest to the community. The types of fatty acids being esterified in membrane phospholipids provide a characteristic fatty acid composition of the phospholipids which can dictate the characteristics of the inflammatory response depending on the types of metabolites of polyunsaturated fatty acids formed through the lipoxygenase (LOX) and cyclooxygenase (COX) pathways, either promoting or inhibiting the inflammatory process, by controlling intracellular signaling pathways, such as protein kinase C (PKC), mitogen-activated protein (MAP) kinases, and phosphoinositol 3 (PI3) kinase [5]. Certain membrane fatty acids also have specific roles in regulation of cell and membrane functions. This is exemplified by gamma linolenic acid (GLA), AA, and EPA which act as precursors for synthesis of an important class of immunoregulatory molecules called eicosanoids. Eicosanoids are a family of 20 carbon-oxygenated derivatives of AA, GLA, and EPA, and include prostaglandins (PGs), thromboxanes (TX), leukotrienes (LTs), and other oxidized derivatives, which are generated from arachidonic acid by the metabolic processes. Eicosanoids are involved in modulating the intensity and duration of inflammatory responses, have cell- and stimulus-specific sources, and frequently have opposing effects. Thus, the overall physiologic (or pathophysiologic) outcome will depend on the cells present, the nature of the stimulus, the timing of eicosanoid generation, the concentrations of different eicosanoids generated, and the sensitivity of the target cells and tissues to the eicosanoids generated [6].

Mechanisms by Which Omega-3 Fatty Acids Influence Inflammation

Polyunsaturated fatty acids (PUFAs) are important constituents of the phospholipids of all cell membranes. They can influence inflammatory cell function and so inflammatory processes by the following ways (Fig. 11.2) [7]:

Altering the Physical Properties of the Membrane

PUFAs can be incorporated into the phospholipids of inflammatory cell membranes where they play important roles assuring the correct environment for membrane protein function, maintaining membrane fluidity, and influencing lipid raft formation [8].

Exerting Effects on Cell Signaling Pathways

It is achieved either through modifying the expression, activity, or avidity of membrane receptors, or through modifying intracellular signal transduction mechanisms that lead to altered transcription factor activity and changes in gene expression. Membrane phospholipids are substrates for the generation of second messengers such as diacylglycerol, and it has been demonstrated that the fatty acid composition of such second messengers, which is determined by that of the precursor phospholipid, can influence their activity [9]. In addition, membrane phospholipids are substrates for the release of (non-esterified) PUFAs intracellularly—the released PUFAs can act as signaling molecules, ligands (or precursors of ligands) for transcription factors, or precursors for biosynthesis of lipid mediators which are involved in regulation of many cell and tissue responses, including aspects of inflammation and immunity.

Altering the Pattern of Lipid Mediators Produced

PUFA intake can influence complex lipid, lipoprotein, metabolite, and hormone concentrations that in turn influence inflammation. Non-esterified PUFAs can act directly on inflammatory cells via surface or intracellular "fatty acid receptors"—the latter may include transcription factors such as peroxisome proliferator-activated receptors (PPARs). PUFAs can be oxidized (enzymatically or non-enzymatically) and the oxidized derivatives can act directly on inflammatory cells via surface or intracellular receptors—oxidation can occur to the non-esterified form of the PUFA or to PUFAs esterified into more complex lipids including circulating or cell membrane phospholipids and intact lipoproteins such as low-density lipoprotein (LDL). The membrane phospholipids of inflammatory cells taken from human-consuming Western-type diets typically contain approximately 10–20 % of fatty acids as arachidonic acid, with about 0.5–1 % EPA and about 2–4 % DHA [10–17], although there are differences between the different phospholipid classes in terms of the content of these fatty acids. The eicosanoid family of inflammatory mediators is generated from 20-carbon polyunsaturated fatty acids (PUFAs) liberated from cell membrane phospholipids. Thus, arachidonic acid is usually the dominant substrate for eicosanoid synthesis. Eicosanoids include PGs, thromboxanes (TXs), leukotrienes (LTs), and hydroxyeicosatetraenoic acids (HETEs). Arachidonic acid in cell membrane phospholipids can be mobilized by various phospholipase enzymes, most notably phospholipase A2, and the free acid can subsequently act as a substrate for the

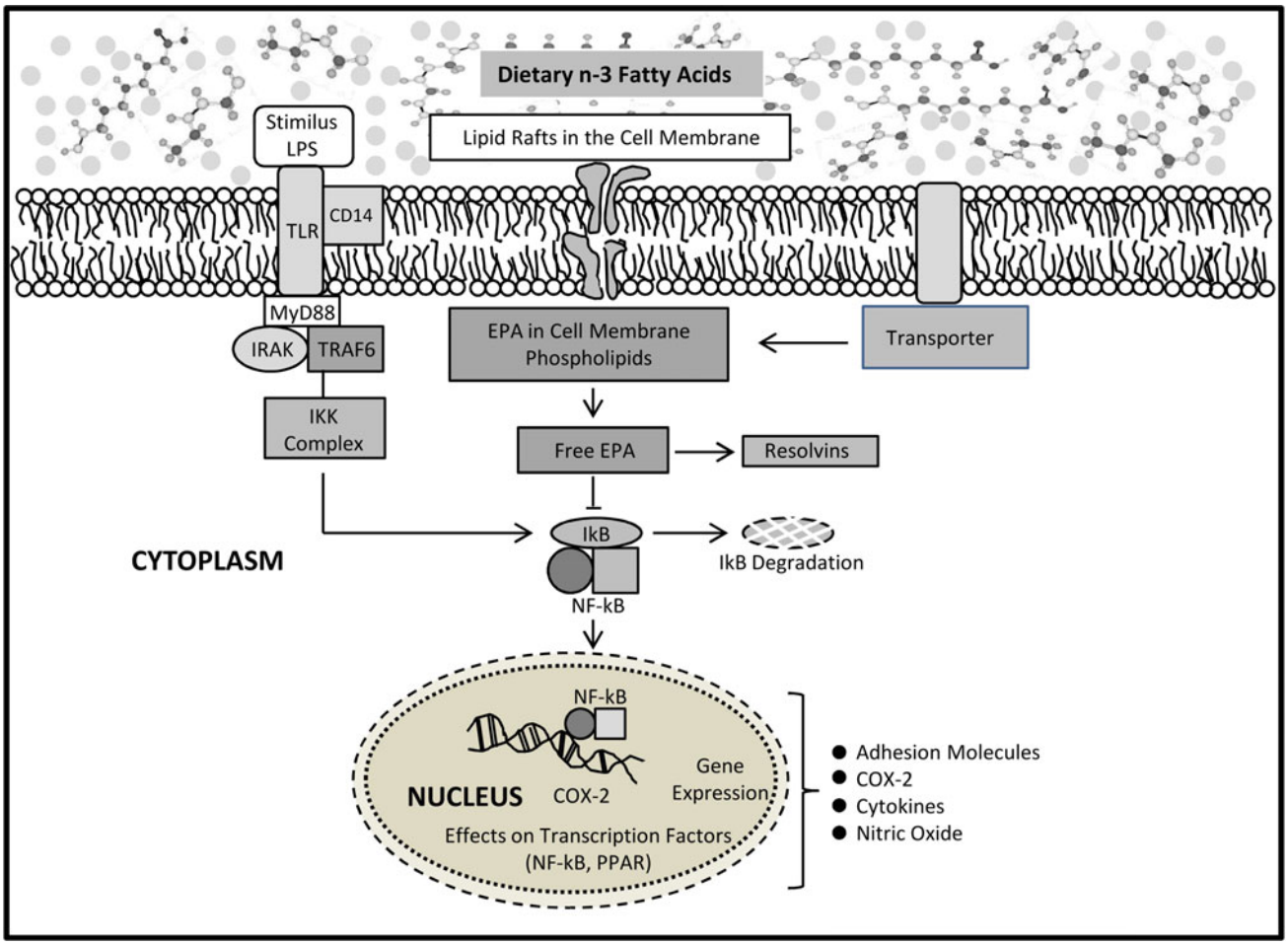

Fig. 11.2 Schematic representation of mechanisms by which omega-3 fatty acids modulate immune response (Modified from [7]) © Springer Science+Business Media, LLC 2010. *COX2* Cycloxygenase2, *EPA* Eicosapentaenoic acid, *LPS* Lipopolysaccharide, *CD14* cluster of differentiation 14, *TLR* Toll-like receptor, *PPAR* Peroxisome proliferator-activated receptor, *NFkB* Nuclear factor kappa B, *IkB* Inhibitory unit of NFkB, *IKK* IkB kinase, *IRAK* Interleukin-1 receptor-associated kinase, *TRAF6* TNF receptor-associated factor 6, *MyD88* Myeloid differentiation primary response gene 88

enzymes that synthesize eicosanoids. Metabolism by cyclooxygenase (COX) enzymes gives rise to the 2-series PGs and TXs. COX-2 is induced in inflammatory cells as a result of stimulation and is responsible for the markedly elevated production of PGs that occurs upon cellular activation. Monocytes and macrophages produce large amounts of PGE2 and PGF2, neutrophils produce moderate amounts of PGE2, and mast cells produce PGD2. Metabolism of arachidonic acid by the 5-lipoxygenase (5-LOX) pathway gives rise to hydroxy and hydroperoxy derivatives (5-HETE and 5-HPETE, respectively), and the 4-series LTs, LTA4, B4, C4, D4, and E4. Neutrophils, monocytes, and macrophages produce LTB4, while LTC4, D4, and E4 tend to be produced by mast cells, basophils, and eosinophils. PGE2 has a number of proinflammatory effects including inducing fever, increasing vascular permeability and vasodilatation, and enhancing pain and edema caused by other agents. PGE2 has been shown to induce COX-2 mRNA expression in cultured fibroblasts and so to upregulate its own production and to induce production of the inflammatory cytokine IL-6 by macrophages [18]. LTB4 increases vascular permeability, is a potent chemotactic agent for leukocytes, induces release of lysosomal enzymes, and enhances generation of reactive oxygen species and production of inflammatory cytokines such as TNF-α, IL-1, and IL-6. The cysteinyl-LTs (LTC4, D4, and E4) are bronchoconstrictors, increase vascular permeability, and promote hypersensitivity. In inflammatory conditions, increased rates of production of arachidonic acid-derived eicosanoids occur and elevated levels of these eicosanoids are observed in blood and tissues from patients with acute and chronic inflammatory conditions. Despite the ongoing emphasis on the proinflammatory effects of arachidonic acid-derived eicosanoids, some of these mediators, for example lipoxin A4, are actually anti-inflammatory [19]. Recent studies have shown that PGE2 inhibits 5-LOX and so decreases the production of inflammatory 4-series LTs and induces 15-LOX promoting the formation of lipoxins that are found to have anti-

Table 11.1 Some diseases with an inflammatory component in which omega-3 fatty acids have beneficial effect

Disease	Conditions
Rheumatoid arthritis	Inflammation of joints
Ulcerative colitis	Inflammation of the mucosa of the colon
Crohn's disease	Inflammation of the ileum and the colon
Asthma	Inflammation of respiratory tract
Multiple sclerosis	Autoimmune disease of brain and spinal cord
Psoriasis	Inflammatory autoimmune disease
Systemic lupus erythematosus	Autoimmune disease affecting any organ system
Chronic obstructive pulmonary disease	Chronic inflammation of the peripheral airways and lung parenchyma
Neurodegenerative disease of aging	Inflammation of central nervous system

inflammatory effects [20]. These findings demonstrate that PGE2 have both proinflammatory and anti-inflammatory actions.

Membrane fluidity and eicosanoid synthesis are the two realms in which lipids have their most potent effects. The effect of dietary fatty acid intake on immune function can be modulated by intake, offering the potential of a dietary management tool in its regulation. Some diseases and conditions that are recognized to having an inflammatory component are listed in Table 11.1. This chapter describes the role of omega-3 fatty acids in rheumatoid arthritis, inflammatory bowel disease, asthma, and multiple sclerosis in detail.

Role of Omega-3 Fatty Acids in Rheumatoid Arthritis

Rheumatoid arthritis (RA) is a chronic, progressive, inflammatory autoimmune disease associated with articular, extra-articular, and systemic effects. The pathobiology of RA is multifaceted and involves T cells, B cells, and the complex interaction of many proinflammatory cytokines TNF-α and IL-6 [21, 22]. The cytokines most directly implicated in this process are TNF-α and IL-6 [20]. These cytokines are messengers that activate and differentiate effector cells that cause local and systemic symptoms associated with this disease.

The cause of rheumatoid arthritis remains unknown, but insights into pathogenic pathways have accumulated over the past two decades [23]. Recent findings suggest a genetic basis for disease development in RA. Environmental factors, such as smoking and infection, may also influence the development, rate of progression, and severity of RA [24, 25]. Various immune modulators (cytokines and effector cells) and signaling pathways are involved in the pathophysiology of RA [22]. The complex interaction of immune modulators is responsible for the joint damage and begins at the synovial membrane [22]. Synovitis is caused by the influx or local activation, or both, of mononuclear cells (including T cells, B cells, plasma cells, dendritic cells, macrophages, and mast cells) and by angiogenesis [22]. The synovial lining then becomes hyperplastic, and the synovial membrane expands and forms villi [22]. The osteoclast-rich portion of the synovial membrane, or pannus, destroys bone, whereas enzymes secreted by neutrophils, synoviocytes, and chondrocytes degrade cartilage [22].

In addition to joint symptoms, many patients experience extra-articular or systemic manifestations, or both [26]. Extra-articular manifestations include rheumatoid nodules, vasculitis, pericarditis, keratoconjunctivitis sicca, uveitis, and rheumatoid lung [26]. Systemic manifestations include acute-phase protein production, anemia, cardiovascular disease (CVD), osteoporosis, fatigue, and depression [27, 28].

Increased understanding of the pathobiology of RA has led to the development of biologic agents that target various immune mediators involved in the disease process [29–42]. Therapies targeted against TNF-α, IL-1, and IL-6, in addition to T- and B cell inhibitors, when used alone or in combination with MTX, have resulted in favorable clinical outcomes in patients with RA [42].

Mode of action of TNF-α inhibitors

TNF-α inhibitors bind with high affinity to soluble and membrane-bound TNF-α and inhibits its effect by blocking TNF-α receptor interactions. It selectively neutralizes membrane-associated and soluble TNF-α and forms high-affinity, stable complexes with soluble and transmembrane bioactive forms of TNF-α, preventing the binding of TNF-α to its receptors [29–38].

Mode of action of other cytokine inhibitors

They neutralize activity of both IL-1a and IL-1b by binding specifically to soluble IL-6 receptor (sIL-6R) and membrane-bound IL-6 receptor (mIL-6R) and inhibiting sIL-6R and mIL-6R-mediated signaling [39, 40]

Mode of action of B- and T- cell inhibitors

B cell inhibitors act by binding CD20 domain expressed on mature B and pre-B cells thereby depleting peripheral B cells temporarily and T cell inhibitors act by selectively blocking the specific binding of receptors of CD80/CD86 on the membrane of the antigen presenting cells with the CD28 receptor on T cells, which is, pathophysiologically, a block of the second signal for activation of T cells. [41, 42].

However, although biologic agents are promising, they are not without limitations [43]. During the 1980s and 1990s, several studies in patients with rheumatoid arthritis showed the beneficial effects of *n-3* PUFA on the development of RA. Several authors reported that fish oils reduce the

Table 11.2 Overview of clinical outcomes in studies using n-3 PUFAs in patients with rheumatoid arthritis

Study and design	Duration of study and no of patients	Placebo	Dose of EPA and DHA (g/d)	Clinical outcomes that improved with intake of n-3 PUFAs	Ref.
DB, PC, P	12 weeks, n = 38	Paraffin oil	1.8 + 1.2	Intake of n-3 PUFAs improved NTJ and DMS	[45]
DB, PC, CO	14 weeks, n = 33	Olive oil	2.7 + 1.8	Intake of n-3 PUFAs improved NTJ, NSJ, TTF and PhyGA	[46]
DB, PC, P	12 weeks, n = 46	Olive oil	3.2 + 2	Intake of n-3 PUFAs improved NTJ and GS	[47]
DB, PC, CO	12 weeks, n = 16	Coconut oil	2 + 1.3	Intake of n-3 PUFAs improved NSJ and DMS	[48]
DB, PC, P	24 weeks, n = 49	Olive oil	Low-dose EPA 1.7 + 1.2 High-dose EPA 3.5 + 2.4	Intake of n-3 PUFAs improved NSJ, NTJ, and GS in low- and high-dose groups and improved DMS and PhyGA in high-dose groups only	[49]
DB, PC, P	12 weeks, n = 27	Coconut oil	2 + 1.3	Intake of n-3 PUFAs improved NSJ and DMS	[50]
DB, PC	24 weeks, n = 43	Mixed oils	1.8 + 1.2	Intake of n-3 PUFAs improved NSJ, NTJ, GS, DMS, and PhyGA	[51]
DB, PC, P	12 weeks, n = 43	Mixed oils	2 + 1.2	Number and severity of tender joints	[52]
DB, PC, P	12 weeks, n = 51	Vegetable oil	2 + 1.2	Intake of n-3 PUFAs improved NTJ, DMS, and CRP	[53]
DB, PC, P	16 weeks, n = 67	Corn oil	3.8 + 1.2	Intake of n-3 PUFAs improved NSJ, STJ, and DMS	[53]
DB, PC	52 weeks, n = 64	Air	1.7 + 1.1	Intake of n-3 PUFAs reduced use of NSAIDs	[54]
DB, PC, P	52 weeks, n = 60	Olive oil	1.7 + 0.4	Intake of n-3 PUFAs improved PtG, and reduced use of NSAIDs	[55]
DB, PC, P	52 weeks, n = 60	Olive oil	0.8 + 0.2	Intake of n-3 PUFAs improved PtG, and reduced use of NSAIDs	[56]
DB, PC, CO	26-30 weeks, n = 49	Corn oil	4.6 + 2.5	Intake of n-3 PUFAs improved NSJ, STJ, PtG, PhyGA, and DMS	[57]
DB, PC	15 weeks, n = 50	Mixed oils	40 mg/kg 2.3 g/d n-3 fatty acids	Intake of n-3 PUFAs improved NSJ, STJ, PtG, HAQ, PhyGA, and DMS	[58]
DB, PC, CO	8 months n = 62	Corn oil	30 mg n-3 fatty acid/kg body wt	Intake of n-3 PUFAs improved NSJ, NTJ, and reduced CRP in those on MTX	[59]
DB, PC, P	16 weeks n = 66	Liquid supplement without added PUFA	1.4 + 0.2 (+0.5 GLA in liquid supplement)	Study did not show superior clinical benefit of daily nutrient supplementation with EPA, GLA at the doses tested as compared to placebo.	[60]
Parallel randomized	24 weeks n = 43	Soybean oil	Total 3 g/d	Intake of n-3 PUFAs improved PtG, JP, GS, RAI, PhyGA, and DMS	[61]
DB, PC,	1 yr, n = 49	Inert oil	240 mg/d EPA with GLA	Intake of GLA with or without EPA reduced use of NSAIDs and improved patient symptoms	[62]

DB Double blind, *PC* Placebo controlled, *CO* Crossover, *P* Parallel, *DHA* Docosahexaenoic acid, *EPA* Eicosapentaenoic acid, *GLA* Gamma Linolenic acid, *HLA* Histocompatibility antigen, *PUFA* Polyunsaturated fatty acid, *LTB4* Leukotriene B4; *NK* Natural killer, *TB3* Thromboxane B3, *NTJ* Number of tender joints, *DMS* Duration of morning stiffness, *NSJ* Number of swollen joints, *TTF* Time to fatigue, *PhyGA* Physician's global assessment, *GS* Grip strength, *CRP* C-Reactive Protein, *PtG* Patient's global assessment, *HAQ* Health assessment by questionnaire, *MTX* Methotrexate, *NSAIDs* Non-steroidal anti-inflammatory drugs, *RAI* Ritchie articular index, *JP* Joint pain

production of inflammatory mediators such as LTB 4 by neutrophils and monocytes [44]. A number of randomized, placebo-controlled, double-blind studies of fish oil treatments for RA have been reported which are listed in Table 11.2 and each concluded the benefit of *n-3* PUFA in RA and suggest that use of n-3 PUFAs as standard therapy for management of RA.

Role of Omega-3 Fatty Acids in Inflammatory Bowel Diseases

Ulcerative colitis (UC) and Crohn's disease (CD) are chronic idiopathic inflammatory disorders of the gastrointestinal tract, collectively termed as inflammatory bowel diseases (IBD). While UC involves exclusively the mucosa of the colon in a variable continuous extent, CD may occur in any part of the digestive tract in a segmental transmural fashion, with the ileum and colon being the most often involved segments [63].

Two transcription factors that are likely to play a role in inflammation of the gastrointestinal tract are nuclear factor kappa B (NFkB) and peroxisome proliferator-activated receptor (PPAR)-γ. NFkB is the principal transcription factor involved in upregulation of inflammatory cytokine, adhesion molecule, and COX-2 genes [64, 65]. NFkB is activated as a result of a signaling cascade triggered by extracellular inflammatory stimuli and involving phosphorylation of an inhibitory subunit (inhibitory subunit of NFkB (IkB)) which then allows translocation of the remaining NFkB dimer to the nucleus [66]. Thus, expression of inflammatory genes is upregulated. NFkB is a recognized target for controlling intestinal inflammation [67–69].

The second transcription factor, PPAR- γ, is also expressed in intestinal tissue [70] where it is believed to act in an anti-inflammatory manner. Colonic biopsies of patients with ulcerative colitis show lowered PPAR-γ expression [71], PPAR-γ knockdown mice show enhanced susceptibility to TNBS-induced colitis [72] and PPAR-γ agonists reduce colitis in murine models [73, 74]. Thus, upregulation of PPAR-γ is also a recognized target for controlling intestinal inflammation [74]. While PPAR-γ directly regulates inflammatory gene expression, it also interferes with the activation of NFkB creating an intriguing interaction between these two transcription factors [75].

There is no curative therapy for these IBDs (except for total proctocolectomy in UC), as its precise etiology remains elusive. IBDs are thought to occur as a result of an inadequate and sustained immune response against luminal (most probably bacterial) antigens, and patients should receive medical treatment for both controlling the inflammatory flares and preventing further bouts of the disease, since they typically have a relapsing and remitting course [76]. Drugs such as aminosalicylates, corticosteroids, immune suppressants (such as thiopurines, cyclosporin, or methotrexate), and biologic agents (mainly anti-TNF monoclonal antibodies) are effective for inducing and/or maintaining remission in IBD [77, 78] but encompass an increased risk for infections and possibility of developing malignancies.

The anti-inflammatory properties of n-3 PUFAs have prompted a series of studies to investigate their efficacy in animal models of inflammatory bowel disease. The primary studies involved chemically induced colitis. The outcomes of these studies are summarized in Table 11.3 and suggest some benefits including improved sigmoidoscopic score, lower relapse rate, and decreased use of corticosteroids. Therefore, dietary management is sought as an alternative approach to IBD therapy.

Role of Omega-3 Fatty Acids in Asthma

Asthma is a chronic inflammatory disorder of the airways leading to airways hyper-responsiveness and associated symptoms such as wheezing and coughing, and is also typically associated with widespread but variable airflow obstruction that is often reversible either spontaneously or with treatment [101]. The inflammatory response is complex and involves a variety of inflammatory cell types including mast cells, alveolar macrophages, neutrophils, eosinophils, lymphocytes, platelets, and a variety of inflammatory mediators [102, 103]. Since airway inflammation is multifactorial, involving various cell types and mediators, the drugs used to decrease inflammation may act at several different steps in the inflammatory process [103, 104]. Various therapeutic strategies have been developed to manage asthma, including the use of short acting beta-2 agonist bronchodilator medications as symptom relievers and anti-inflammatory preventer medications such as inhaled corticosteroids and oral leukotriene antagonists [103, 104]. While pharmacological medications have proven highly effective and have facilitated the management of asthma, prolonged use of some medications may result in reduced efficacy or tachyphylaxis [105, 106]. There is accumulating evidence that dietary modifications have the potential to influence the severity of asthma and reduce the dose requirements of drug treatment. Therefore, various studies to relate the effect of n-3 PUFA supplementation on patients suffering from bronchial asthma have been conducted and have demonstrated different levels of benefit. Though there are inconsistency among study results which may be attributed to the heterogeneity in definitions of the study populations (e.g., age, gender, clinical picture of asthma including its severity), and the type of intervention (e.g., amounts of oil and omega-3 fatty acid contents). Only few data are available on the effect of n-3 PUFA supplementation on patients with asthma which are listed in Table 11.4.

Table 11.3 Overview of clinical outcomes in studies using n-3 PUFAs in patients with inflammatory bowel disease

Disease (Study and design)	Duration of study and no. of patients	Placebo	Dose of EPA and DHA (g/d)	Clinical outcomes that improved with intake of n-3 PUFAs	Ref.
Ulcerative colitis (DB, PC, CO)	4 months n = 11	Mixed oils (Oleic + Palmitic + Linoleic)	2·7 + 1·8	56 % mean reduction in DAI with fish oil versus 4 % with placebo, decreased use of corticosteroids	[79]
Ulcerative colitis (DB, PC, CO)	4 months n = 18	Mixed oils (Oleic + Palmitic + Linoleic)	3·2 + 2·2	Greater improvement in histology index with fish oil (P = 0·002). No differences in clinical and endoscopic response	[80]
Ulcerative colitis (DB, PC, P)	4 months n = 53	Sunflower oil	1·5 g EPA/day + 2·1 g GLA/day	n-3 and n-6 (alone or in combination) had no steroid sparing effect.	[81]
Ulcerative colitis (DB, PC, P)	6 months n = 18	Sunflower oil	3·2 + 2·4	Improvement in clinical score with fish oil ($p < 0·05$) but not with placebo. Lower endoscopic and histological scores at 6 months with fish oil	[82]
Sulfasalazine-controlled, crossover study	2 months treatment and 2 months washout n = 10	Sulfasalazine 2 g per day	3·2 + 2·2	Increase in C-reactive protein, ESR and platelet count in the fish oil group. In spite of that endoscopic score improved with fish oil	[83]
Ulcerative colitis (DB, PC, P)	6 months n = 51	Sunflower oil	5·6 g n-3 PUFA/day	Clinical and endoscopic improvement in the EFA group	[84]
Ulcerative colitis (DB, PC, P)	6 months n = 121	Liquid supplement based on sucrose alone	Nutritional liquid supplement with <2·5 g EPA and <1·0 g DHA per day (plus prebiotics and antioxidant micronutrients	Improvement in clinical, endoscopic and histological indices. Faster reduction in steroid dose with active therapy	[85]
Ulcerative colitis (DB, PC, P)	12 months n = 87	20 ml/d of olive oil	20 ml/d (5·0 g EPA, + 2·1 g DHA)	EPA decreased serum leukotriene B4 levels, reduced the need for corticosteroids but only modestly improved clinical parameters	[86]
Ulcerative colitis (DB, PC, P)	24 months n = 64	Corn oil	5·1 g n-3 PUFA per day	n-3 FAs demonstrated little long-term benefits as assessed by colonoscopy, histology	[87]
Ulcerative colitis (DB, PC, P)	12 months n = 50	20 ml/day olive oil	20 ml/day (3·2 g EPA + 2·2 g DHA)	No effect on relapse rate.	[88]
Ulcerative colitis (DB, PC, P)	12 months n = 63	Sunflower oil	EFA capsules: 1·62 g GLA*, 0·27 g EPA, 0·05 g DHA per day	55 % relapse rate with EFA versus 38 % with placebo	[89]
Crohn's disease (DB, PC, P)	9 weeks n = 31	Nutritional supplement with 7·8 g Linoleic acid (n-6 FA) per day	Nutritional supplement enriched with 3·0 g n-3 FA (EPA, DHA, ALA), 11·4 g L-Arginine, and 1·2 g RNA per day	Significant decreases in CDAI and C-reactive protein in active and control groups	[90]

(continued)

Table 11.3 (continued)

Disease (Study and design)	Duration of study and no. of patients	Placebo	Dose of EPA and DHA (g/d)	Clinical outcomes that improved with intake of n-3 PUFAs	Ref.
Crohn's disease (DB, PC, P)	9 weeks n = 41	Elemental diet with 0·4 % of energy as n-3 FA (ALA) and 5·4 % as n-6 FA	Polymeric diet with 1·5 % of energy as n-3 FA (ALA) and 3 % as n-6 FA (LA)	71 % remission rate with active versus 70 % with control formula	[91]
Crohn's disease (DB, PC, P)	12 weeks n = 78	Enteric-coated MCT capsules with 2·7 g caprylic and 1·8 g capric acid per day	Enteric-coated capsules with 1·8 g EPA/day + 0·9 g DHA/day	28 % relapse rate with fish oil versus 69 % with placebo	[92]
Crohn's disease (DB, PC, P)	12 weeks n = 135	Corn oil (All patients on a high-fiber, low arachidonic acid diet)	Fish oil capsules: Omega-3 PUFA (3·3 g EPA and 1·8 g DHA per day) as ethyl esters	57 % relapse rate with fish oil versus 55 % with placebo	[93]
Crohn's disease (DB, PC, P)	12 months n = 50	Not described	Fish oil capsules: Enteric-coated capsules with 1·8 g EPA/day and 0·9 g DHA/day	8 % clinical recurrence rate with fish oil versus 21 % with placebo	[94]
Crohn's disease (DB, PC, P)	12 months n = 38 pediatric patients	Olive oil	Fish oil capsules: Enteric-coated capsules with 1·2 g EPA and 0·6 g DHA per day	61 % relapse rate with fish oil versus 95 % with placebo	[94]
Crohn's disease (DB, PC, P)	52 weeks n = 363 adult patients	Enteric-coated capsules with MCT (4 g/d)	Enteric-coated capsules with 2·0–2·4 g EPA and 0·6–1·0 g DHA per day	31·6 % relapse rate with fish oil versus 35·7 % with placebo	[95]
Crohn's disease (DB, PC, P)	52 weeks n = 375 adult patients	Enteric-coated capsules with MCT (4 g/d)	Enteric-coated capsules with 2·0–2·4 g EPA and 0·6–1·0 g DHA/day	47·8 % relapse rate with fish oil versus 48·8 % with placebo	[96]
Ulcerative colitis	12 weeks n = 6	Not described	3–4 g/d EPA	Lower levels of LTB4	[97]
Ulcerative colitis	8 weeks n = 10	Not described	2.7 g/d EPA 1.8 g/d DHA	Amelioration of disease markers	[98]
Crohn's disease	2 years n = 38	Not described	100–250 g fish	Remissión of major symptoms	[99]
Ulcerative Colitis (PC)	6 months n = 43	Primrose oil/olive oil	Max-EPA	EPA produced minimal changes in clinical outcome as assessed by sigmoidoscopy, rectal biopsy, and symptoms	[100]

DB Double blind, *PC* Placebo controlled, *CO* Crossover, *P* Parallel, *DHA* Docosahexaenoic acid, *EPA* Eicosapentaenoic acid, *GLA* Gamma Linolenic acid, *PUFA* Polyunsaturated fatty acid, *LTB4* Leukotriene B4; *DAI* Disease activity index, *ESR* Erythrocyte sedimentation rate, *TB3* Thromboxane B3, *NSAIDs* Non-steroidal anti-inflammatory drugs *MCT* Medium chain triglycerides, *CDAI* Crohn's disease activity index

Role of Omega-3 Fatty Acids in Multiple Sclerosis

Multiple sclerosis (MS) is a chronic inflammatory disease of the central nervous system (CNS). The target cells in the pathogenesis of MS are oligodendrocytes, the myelin-forming cells of the CNS. At present, the cause of onset of MS is unknown, but activated T cells and macrophages are thought to be involved in demyelination through various mechanisms. The important pathological mechanisms involved in MS include immune-mediated inflammation [117], oxidative stress [118–120], and excitotoxicity [121]. These mechanisms

Table 11.4 Overview of clinical outcomes in studies using n-3 PUFAs in patients with asthma

Study and design	Duration of study and no. of patients	Placebo	Dose of EPA and DHA (g/d)	Clinical outcomes that improved with n-3 PUFA supplementation	Ref.
Parallel RCT	10 week n = NR	Olive oil placebo	3.2 g/d EPA + 2.2 g/d DHA	Improved PEK	[107]
Parallel RCT	8 week n = 23	600 mg/d olive oil	200 mg/d EPA/DHA + 400 mg/d	There was a significant decrease in daytime wheeze, the concentration of exhaled H_2O_2 and an increase in morning PEF in the lipid extract group compared to the placebo group	[108]
Parallel RCT	8 week (n = 6)	0.1 g/d EPA ethyl ester (+trace DHA)	4.0 g/d EPA ethyl ester (+trace DHA)	None identified	[109]
Crossover RCT	10 week (n = 15)	15 g/d olive oil placebo	2.7 g/d EPA + 1.8 g/d DHA	None identified	[110]
RCT	4 week (n = 7)	10–20 g/d corn oil	10–20 g/d Perilla seed oil (ALA: NR)	Supplementation of Perilla seed oil-rich diet suppressed generation of LTB4 and LTC4 by leukocytes and improvement of pulmonary function	[111]
Crossover RCT	10 week (n = 36)	20 ml/d olive oil/20 ml/d evening primrose oil	20 ml/d fish oil (EPA + DHA)	Moderate doses of evening primrose oil or fish oil are ineffective as a supplementary treatment of bronchial asthma	[112]
Parallel RCT	26 week (n = NR)	Olive oil placebo (dose: NR)	3.2 g/d EPA + 2.2 g/d DHA	None identified	[113]
Parallel RCT	26 week (n = NR)	n-6: 1.8 g/d safflower oil + 1.8 g/d palm oil + 0.4 g/d olive oil + sunflower diet (dose: NR)	0.72 g/d EPA + 0.48 g/d DHA + ALA (dose: NR) via canola diet	None identified	[114]
Parallel RCT	10 month (n = 15)	300 mg/d olive oil placebo (n = 15)	17.0–26.8 mg/kg/d EPA; 7.3–11.5 mg/kg/d DHA (300 mg/d fish oil)	Decreased asthma symptom scores; decreased bronchial hyper-responsiveness to acetyl choline challenge	[115]
Parallel RCT	9 month (n = NR)	"Placebo" (type and dose: NR)	1.0 g/d EPA + DHA	Improved FEV_1	[116]

DB Double blind, *PC* Placebo controlled, *CO* Crossover, *P* Parallel, *RCT* Randomized controlled trials, *DHA* Docosahexaenoic acid, *EPA* Eicosapentaenoic acid, *ALA* Alpha-linolenic acid, *GLA* Gamma-linolenic acid, *PUFA* Polyunsaturated fatty acid, *LTB4* Leukotriene B4, *LTB4* Leukotriene C4, *TB3* Thromboxane B3, *NR* Not reported; *FEV_1* Forced expiratory volume at 1 s, *PEK* Peak expiratory flow

may all contribute to oligodendrocyte and neuronal damage and even cell death, hence promoting disease progression.

At present, no therapy exists that can confer prolonged remission in MS and therapeutic agents are only partially effective. Their long-term beneficial effects are uncertain and often detrimental side effects have been reported [122, 123]. In a recent survey, 37 % of 1,573 patients with MS revealed that they had used omega-3 unsaturated fatty acids at some point in their lives [124]. Several small studies have demonstrated a reduction in PUFA content in serum, cerebral white matter, erythrocytes, and lymphocytes in patients with MS compared with controls [125–128]. However, these observations do not help to clarify the exact nature of the relationship between PUFA intake and MS, as no data were provided on the dietary habits and clinical characteristics of the study participants.

In an attempt to provide a proper assessment of the efficacy of PUFA supplementation in MS, multiple controlled studies have been performed, some of which date back to the 1970s. These studies, however, generally produced inconclusive results. The results of the controlled trials performed to date are summarized in Table 11.5.

Summary and Conclusion

Inflammation is the root cause of a number of degenerative diseases such as rheumatoid arthritis, inflammatory bowel disease, asthma, multiple sclerosis, and atherosclerosis. Although steroidal anti-inflammatory drugs (SAID) and non-steroidal anti-inflammatory drugs (NSAIDs) are used effectively to manage the acute inflammatory reaction, their

Table 11.5 Overview of clinical outcomes in studies using n-3 PUFAs in patients with multiple sclerosis

Study and design	Duration of study and no. of patients	Dietary	Dose of EPA and DHA (g/d)	Clinical outcomes	Ref.
DB, P	24 months n = 87 patients with DSS scores from 0–6	Oleic acid (7.6 g/day) emulsion	Linoleic acid (17.2 g/day) emulsion	Significant improvement in relapse severity and nonsignificant trend toward lower annualized relapse rates in the linoleic acid group; no differences in disability between the two groups	[129]
DB, P	24 months n = 152	Oleic acid (4.8 ml/day)capsules and oleic acid (4 g/day) spread	Four treatment arms: linoleic acid (0.36 g/day) + GLA (3.42 g/day) capsules and linoleic acid (11.5 g/day) spread	No significant differences in disability (measured on the DSS), relapse rates or relapse severity score among the four groups	[130]
DB, P	24 months N = 116 patients with relapsing MS	Oleic acid (4.0 g/day) capsules and oleic acid (16 g/day) spread	Four treatment arms: linoleic acid (0.34 g per day) + GLA (2.92 g/day) capsules and linoleic acid (23 g/day) spread	Linoleic acid plus linolenic acid group had briefer and less-severe relapses compared with placebo group, but accumulated more disability than placebo group	[131]
DB, P	30 months n = 96 patients with relapsing and progressive MS	Oleic acid 21 g/day	Linoleic acid 17 g/day	No differences in disability, rates, or severity of relapse, or timed functional tests between the two groups; significant increase in serum concentrations of linoleic acid in the active arm	[132]
DB, P	24 months, n = 312 patients with relapsing MS	Oleic acid (7.2 g/day) capsules	Fish oil (mixture of EPA 1.71 g/day and DHA 1.14 g/day) capsules	Fish oil group showed a nonsignificant trend toward less disability progression	[133]
DB, P	12 months n = 31 patients with relapsing MS	Oleic acid (1.0 g/day) capsules	Fish oil (EPA 1.98 g/day and DHA 1.32 g/day) capsules	No differences seen in relapse rates between the two groups; fish oil group had improvements in quality-of-life measures	[134]
DB, P	18 months n = 36 patients with active MS	Polyethylene glycol	High-dose GLA (14 g/day) versus low-dose GLA (5 g/day)	High-dose GLA group had significantly reduced relapse rates and disability progression (measured on the expanded DSS) compared with low-dose GLA and placebo groups	[135]

DB Double blind, *P* Parallel, RCT, *DHA* Docosahexaenoic acid, *EPA* Eicosapentaenoic acid, *ALA* Alpha-linolenic acid, *GLA* Gamma-linolenic acid, *PUFA* Polyunsaturated fatty acid, *LTB4* Leukotriene B4, *LTB4* Leukotriene C4, *TB3* Thromboxane B3, *NR* Not reported, *MS* Multiple Sclerosis, *DSS* Disability Status Scale

use for chronic inflammation is followed by severe adverse effects. This has given an impetus to search for alternate natural and safe anti-inflammatory agents.

The knowledge that dietary nutrients can act as drugs for ameliorating disease captured attention of researchers to figure out the active component of the particular dietary source responsible for protective effect. Population studies revealed the anti-inflammatory and cardioprotective effects of omega-3 fatty acids, with subsequent clinical studies (prospective randomized placebo-controlled trials) supporting their therapeutic role in chronic inflammatory conditions such as rheumatoid arthritis, inflammatory bowel disease, asthma, and cardiovascular disease.

Omega-3 fatty acids act by increasing production of anti-inflammatory eicosanoids and inflammation resolving resolvins from EPA and DHA, downregulating adhesion molecule expression on leukocytes and on endothelial cells, reducing intercellular adhesive interactions and production of proinflammatory cytokines induced via the NFkB system and decreasing chemotactic responses of leukocytes.

The supplementation trials in patients with rheumatoid arthritis appear to be the most successful with most trials reporting several clinical benefits. In most other inflammatory diseases and conditions, there are either too few studies or unequivocal results to draw a clear conclusion of the possible efficacy of omega-3 fatty acids as a treatment. Hence, additional studies are needed to conclude about the effective dosage and duration of omega-3 fatty acid administration for best possible clinical benefit in a particular inflammatory condition.

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