Pain

Patients' most common complaint & the 1st one they want fixed!

Disclosures: none

Objectives:

1. Review the anatomy & physiology of pain
2. To understand the similarities of very divergent causes of pain
3. What are the possible therapies prior to the use of controlled substances

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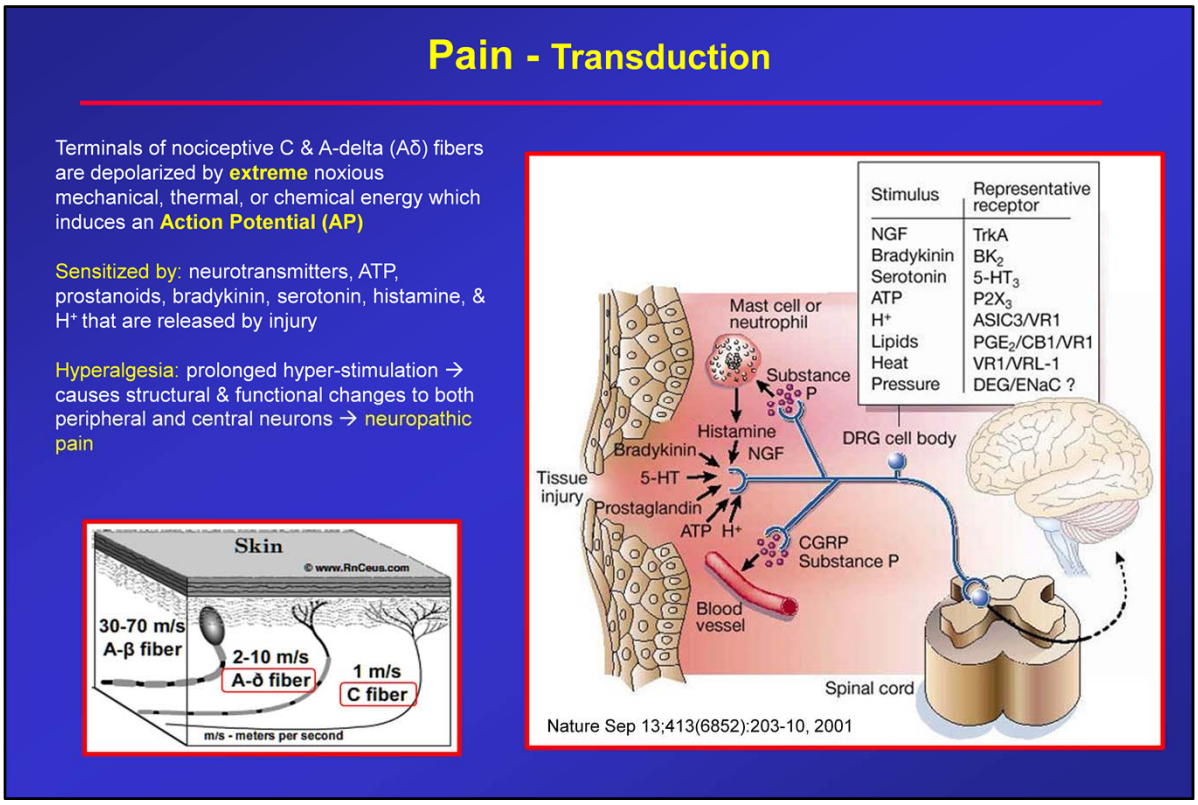
Pain

General approach to pain therapy:

1. Fix the cause
2. Block the signal from reaching the brain
3. Increase the activity of the inhibitory pathways
4. Use cortical functions to help alleviate the patient's pain perception

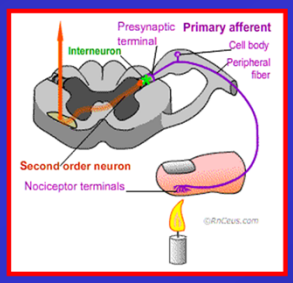


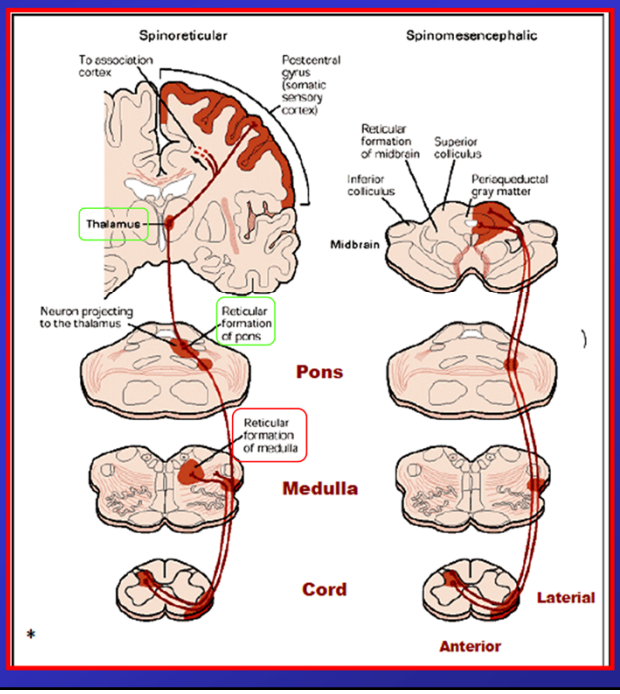
Transduction begins when peripheral terminals of nociceptive C fibers and A-delta (Aδ) fibers are **depolarized by noxious mechanical, thermal, or chemical energy.** The membranes of these terminals contain **proteins and voltage-gated ion channels** that convert thermal, mechanical, or chemical energy into an **action potential (AP).** Nociceptor terminals are spread densely throughout the skin. They are found less on periosteum, joints, tendons, muscles, and least on the surface of organs. Normally, nociceptor terminals have a **high activation threshold**. They require an intense stimulation to generate an AP. For example, thermal nociceptors are only activated by temperature extremes (>45°C or < 5°C). However, nociceptors can be **made more sensitive to stimuli**. Injury to neurons and surrounding tissues expose neighboring nociceptors to irritating substances, including: neurotransmitters, ATP, prostanoids, bradykinin, serotonin, histamine, and hydrogen ions (acid pH), etc. These substances lower the nociceptor's activation threshold (**sensitize**), creating a condition of hyperesthesia. Hyperesthesia is a term that encompasses both allodynia and hyperalgesia. A common example of allodynia is the painful response to touch in an area of a 1st degree burn, e.g. sunburn. Normally, allodynia subsides as healing progresses. Hyperalgesia, on the other hand, results from prolonged hyper-stimulation which can cause structural and functional changes to both peripheral and central neurons. These changes can cause central sensitization leading to the development neuropathic pain.

Pain – Conduction & Transmission

AP generated in NC terminals is conducted through peripheral nerve to dorsal horn of spine → depolarizes presynaptic terminal.
Presynaptic terminal interfaces with network of interneurons & second order neurons.
(Release **Glutamate, substance P, & CGRP**)

Spino-reticular connect to **medulla** & **cortex**
(Synapses in Pons & Thalamus)
Spino-mesencephalis to **midbrain**
Many neurotransmitters released
NMDA channels are blocked by **Mg** but excess stim releases Mg & allows release of nitric oxide → exaggerated release of transmitters





Conduction of an AP is the second phase of nociception. An AP generated in nociceptor terminals is conducted across the peripheral process to the central process were it depolarizes the presynaptic terminal. The presynaptic terminal interfaces with a network of interneurons and second order neurons in the dorsal horn. Interneurons can facilitate or inhibit transmission to second order neurons. There are 2 types of nociceptor fibers that conduct APs to the spinal cord. A-delta fibers (Að) are slow, thin, myelinated fibers associated with sharp/pricking, well localized pain. C fibers are very slow, thin, unmyelinated fibers that are associated with a dull, aching, throbbing, diffuse pain. A third type of fiber, A-beta(Aß), is a fast, large diameter, myelinated fiber that carries APs from mechano-receptors. Aß fibers are believed to modulate C and Að activity within the dorsal horn.

Transmission: The **antero-lateral white matter** of the cord contains not only the spino-thalamic tract, but other pathways arising from the dorsal horn and known to carry nociceptive signals. Some of these axons project to the reticular formation and the midbrain, and together they form the **Anterolateral System**. Some axons in the pain pathway project to the reticular formation (spino-reticular fibres) and to the peri-aqueductal grey matter of the midbrain.

Alternative Pain Pathways in the Antero-lateral System
Nociceptive signals are distributed not only to the thalamus (via the spino-thalamic

tract), but also to areas of the reticular formation of the brainstem, and to the periaqueductal grey matter of the midbrain. All of these pathways originate within the dorsal horn and cross the midline near the central canal.

Spino-Reticular Tracts

The reticular formation of the brainstem is a hotchpotch of neurones that tend not to be grouped into well defined nuclei. They re the substance of the brainstem which surrounds the identifiable nuclei (such as the cranial nerve nuclei) and the fibre tracts thatcourse through the pons and medulla. Some groups of neurones within distinct boundaries are given names, such as the nucleus gigantocellularis reticularis and the midline raphe nuclei. The left hand diagram shows the spino-reticular tract, whose axons terminate in the medulla and pons. Like the spino-thalamic tract these axons ascend the cord in the antero-lateral quadrant. The axon terminals synapse in many parts of the brainstem and the third order neurons of the pathways project to the thalamus.

The third phase begins when a nociceptive AP reaches the presynaptic terminal in the dorsal horn. The AP causes the presynaptic terminals of Að and C fibres to release a variety of pro-nociceptive substances into the synaptic cleft. C-fiber presynaptic terminals are known to release **glutamate** which activates postsynaptic α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid **(AMPA) receptors**; **substance P (SP)**, which activates postsynaptic **NK1 receptors**; and **calcitonin gene–related peptide (CGRP),** which activates postsynaptic **CGRP receptors**. Activation of postsynaptic receptors results in an influx of ions that depolarize second order neurons and interneurons. When a secondary neuron is depolarized it generates an action potential that is relayed through the contralateral spinothalamic tract (STT) to the medulla and brain stem via the spinoreticular (SRT) & spinomesencephalic (SMT) tracts or to the hypothalamus via the spinohypothalamic tract (SHT). Postsynaptic terminals of second order neurons exhibit an array of neurotransmitter receptors. An unusual and important receptor is the N-methyl-D-aspartate **(NMDA)-linked channels**. NMDA channels are normally inactive because they are usually blocked by magnesium ions. However, intense or prolonged periods of depolarization can release the magnesium ion from the NMDA-linked channel. Loss of the Mg blockade allows an influx of calcium ions. Increased intracellular calcium ions has two important effects on the development of neuropathic disease:

The first effect leads to a lowered activation threshold, a change in cation channel kinetics, and the insertion of more receptors in the postsynaptic membrane.

The second effect initiates a cascade that results in the production and release of **nitric oxide** into the synaptic cleft. Presence of nitric oxide in the synaptic cleft causes an

exaggerated release of neurotransmitters from the presynaptic terminal resulting in synaptic hyperexcitability.

Pain – Visceral

Visceral pain:
One of the strongest painful sensations known to man caused by ischemia, layer friction, mesenteric stretch, organ distension, or inflammation.

Spinal afferents that accompany the autonomic nerves are responsible for initiating visceral pain.

Second order neurons that project to brain also carry afferent traffic from the skin.

Caused by viscero-somatic convergence → both pathways (visceral and somatic) converge on the same neurons in the dorsal horn.

Activity in visceral afferents can be confused with somatic inputs within the same segments, and perceived as sensations within those somatic areas.

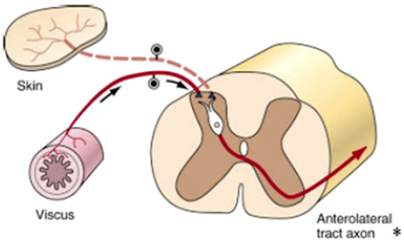
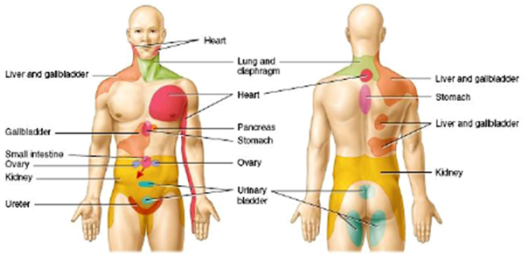


Diagram of the convergence of somatic and visceral afferents on the neurones of the anterolteral system.



Referred Pain : Visceral Pain

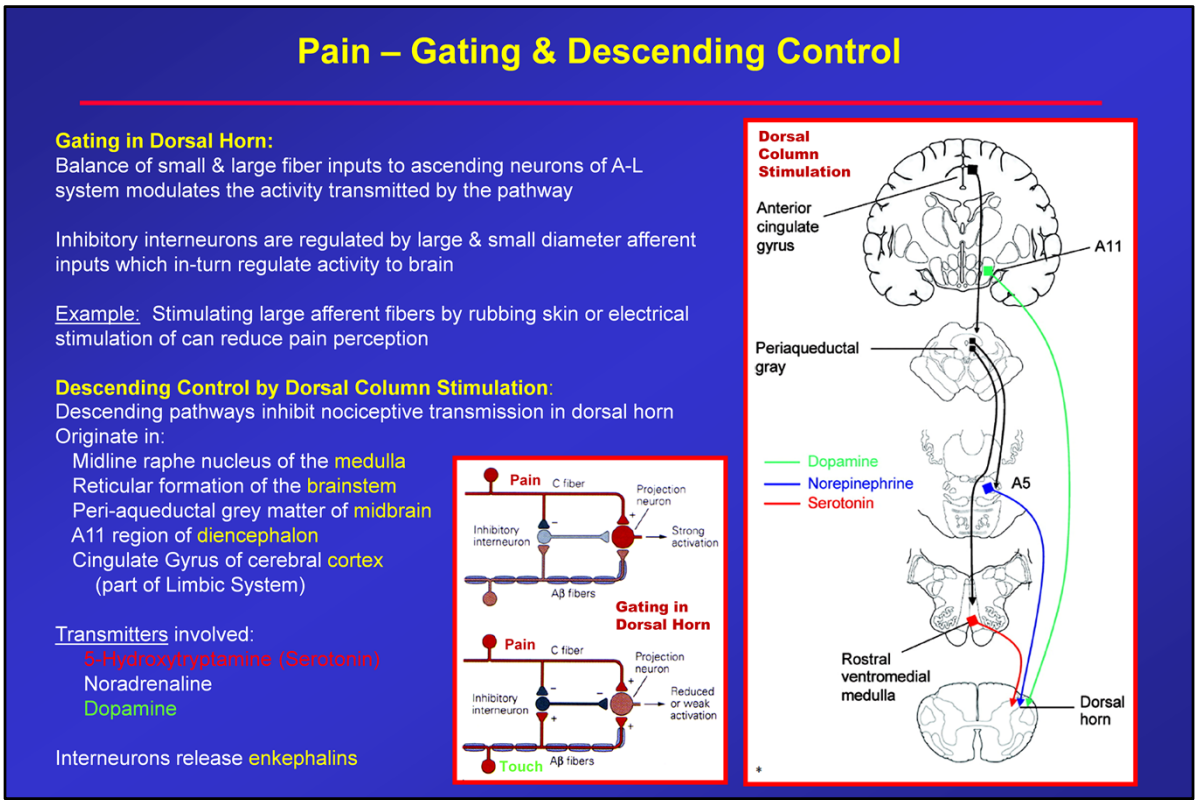
Visceral pain is one of the strongest painful sensations known to man. It arises from internal organs as a result of occlusion of arteries (as in a heart attack), friction between layers of pleura or peritoneum (as in pleurisy or rebound tenderness in the abdomen), stretch of mesenteries, over distension of viscera, or inflammation.

The spinal afferents that accompany the autonomic nerves are responsible for initiating visceral pain, but the second order neurons that project rostrally also carry afferent traffic from the skin. This is because of viscero-somatic convergence - both pathways (visceral and somatic) converge on the same neurons in the dorsal horn. A consequence of this is that activity in visceral afferents can be confused with somatic inputs within the same segments, and perceived as sensations within those somatic areas.

Some examples:
Afferents from the heart can induce sensations referred to the chest and arm (and sometimes in the neck)
Afferents from the gall bladder can induce sensations in the abdominal skin, but also in the shoulder (because inflammation in that region can activate afferents of the phrenic nerve, whose origin is C4-5, which also innervates the shoulder)
Afferents from the kidney and ureter can give rise to pain radiating from the small of

the back to the groin.

Tractotomy (Cordotomy). Neurosurgeons sometimes section the anterolateral system in an attempt to relieve chronic intractable pain by dividing the axons in the spinal cord. This procedure can be successful, but breakthrough pain sensation can occur after a year or so, and one reason for this may be that there are alternative ascending pain pathways in the anterolateral system and other smaller pathways (such as the post-synaptic dorsal column pathway).



The schematic diagram above shows the concept of 'gating' in the dorsal horn of the spinal cord. It hypothesizes that the balance of small and large fiber inputs to ascending neurons of the antero-lateral system modulates the activity transmitted by the ascending pathway.

The diagrams show a projection neuron, possibly equated with the marginal cells of Lamina I that project to the thalamus in the anterolateral system, and the arrangement of afferent neurons that could alter transmission in that pathway.

An inhibitory interneuron is shown (possibly equating to a Lamina II neuron) and the large and small diameter afferent inputs that regulate the activity of that cell can be seen.

The top diagram shows the response to stimulation of small fibers (nociceptors), whereas the bottom indicates the response to simultaneous activation of tactile and nociceptors.

The conclusion is that circuits of this type can modulate the activity of the antero-lateral system, depending on the balance of small and large diameter afferent inputs.

The **Gate Theory** suggests that the perception of pain depends on the balance of large and small fiber activity entering the dorsal horn, and that large myelinated fiber activity can reduce the onward transmission of nociceptive signals carried by unmyelinated axons.

There is now a lot of evidence that the pain signal can be modulated in the dorsal horn. Stimulation of large afferent fibers, by rubbing the skin, or by electrical stimulation of the skin (**TENS**) can reduce the perception of pain, and it is a common behavior to rub an area of skin that has recently been hit by a hard object.

Dorsal column stimulation has been performed in humans using implanted electrodes; these stimulate a collateral of the large afferent neurons, and the impulse that passes antidromically down the cord appears to be able to reduce the perception of pain in patients with chronic pain conditions. The suggestion is that this mechanism is identical to that proposed in the Gate Theory.

Descending Control :
It is believed that the synapses in the dorsal horn can modulate the pain message. This can happen through to main mechanisms:
interactions between innocuous and nociceptive inputs to the dorsal horn (the 'Gate' Theory), and descending pathways that terminate in the dorsal horn.

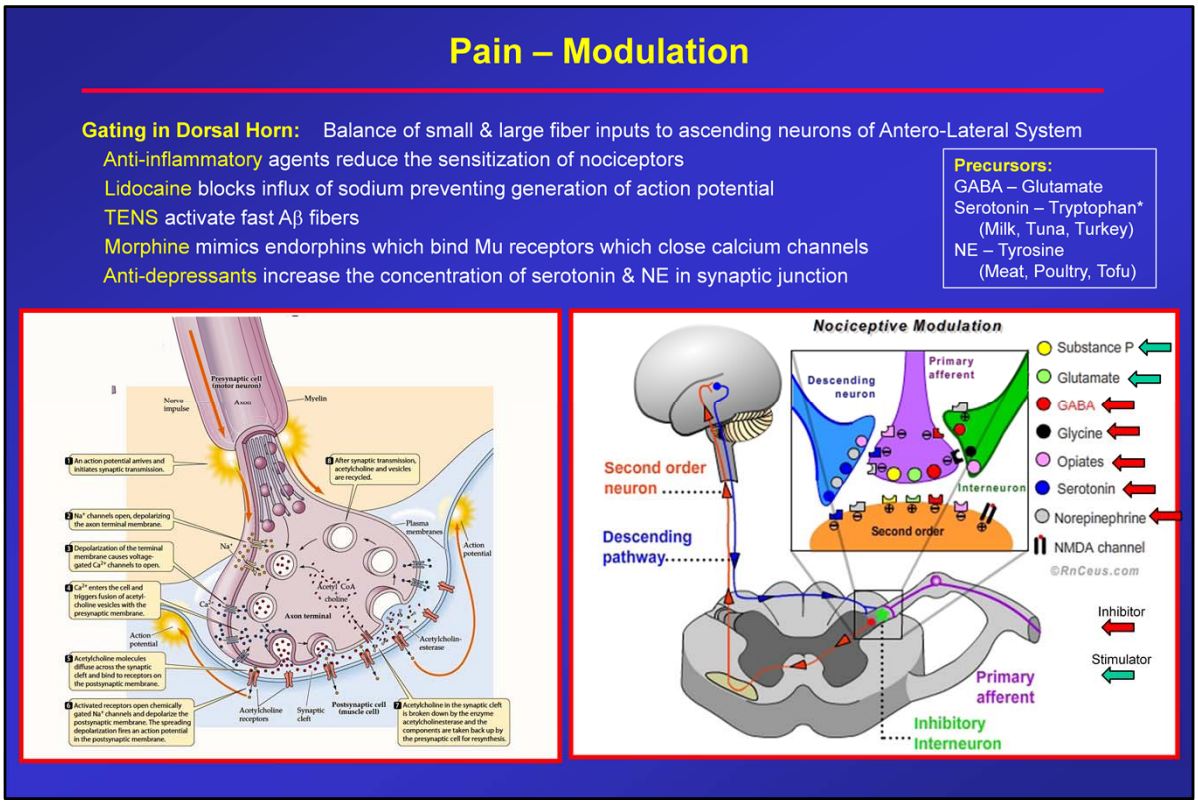
The diagram shows some of the descending pathways that, when stimulated, inhibit nociceptive transmission in the dorsal horn. They originate in:

- midline raphe nucleus of the medulla
- reticular formation of the brainstem
- peri-aqueductal grey matter of the midbrain
- region of the diencephalon known as A11
- Cingulate Gyrus of the cerebral cortex (part of the Limbic System)

The diagram also shows the importance of **amines** in influencing nociceptive processing in the dorsal horn. Several transmitters are involved in the process:

- 5-Hydroxytryptamine (Serotonin)
- Noradrenaline
- Dopamine

In addition there are interneurons in the dorsal horn that release enkephalins, which also have an inhibitory action at this site.



Modulation of nociceptive transmission is an adaptive process involving both excitatory and inhibitory mechanisms. In the normally functioning state, the responses of second order neurons can be suppressed or facilitated dependent on other events important to the organism. For our purposes, we will focus on the processes that inhibit or suppress transduction, conduction, or transmission, thereby interrupting or diminishing the perception of pain.

Peripheral modulation can be accomplished by:
a) Inhibiting the sensitization of **nociceptor terminals** with medications such as cyclooxygenase inhibitors, e.g. aspirin, ibuprofen, etc.

b) Inhibiting depolarization and repolarization of the axonal membrane. Local anesthetics like **lidocaine** prevent the generation or conduction of an action potential by blocking the **influx of sodium** through voltage-gated sodium channels located along first and second order afferents.

c) Inhibiting the **inflammatory response** to trauma with hydrocortisone. Hydrocortisone is believed to stimulate the production of **lipocortin-1**, which inhibits the biosynthesis of prostaglandins and leukotrienes from arachidonic acid by inhibiting the cytosolic enzyme phospholipase A2. Lipocortin-1 is also believed to inhibit leukocytic inflammatory events including: epithelial adhesion, emigration,

hemotaxis, phagocytosis.

d) Stimulating the large fast Aβ fibers in the area of injury can induce interneurons in the dorsal horn to release **GABA** and **glycine** which inhibit the release of **glutamate** from the primary afferent terminal, thereby preventing depolarization of the second order neuron. Mechanical stimulation and transcutaneous electrical nerve stimulation (TENS) are believed to reduce the perception of pain by activating fast Aβ fibers.

Central modulation of nociception involves multiple sites and mechanisms:
a) Exogenous opioids like hydromorphone, morphine and oxycodone produce analgesia by mimicking endogenous **endorphins**. Opioids are able to activate the endorphin receptors: Mu, Kappa and Delta. **Mu receptors** are responsible for most of the analgesic effect of opioids and are present on neurons in the spinal cord, brainstem, and midbrain.

b) In the dorsal horn, Mu receptor act by closing voltage sensitive calcium channels on the primary afferent presynaptic terminal. Blocking the influx of Ca^{++} inhibits depolarization and the subsequent release of the neurotransmitters **glutamate** and **substance P**. Mu receptors also increase the efflux of K^{+} from the second order postsynaptic terminal, which increases the internal (-) charge, creating a hyperpolarized state. In the brainstem and midbrain, activated Mu receptors turnoff GABAergic interneurons responsible for suppressing the anti-nociceptive descending pathway. In other words, opioids enhance the activity of the descending pathway which results in increased release of anti-nociceptive **serotonin** and **norepinephrine** from the descending neuron terminals into the dorsal horn.

b) Endogenous opioids (**endorphins**) work in a manner similar to exogenous opioids. The existence of endorphins has been demonstrated by the application of stimulation that produces analgesia (SPA). Electrical stimulation of the periaqueductal gray (PAG) elicits the release of endorphins which produce an analgesia that can be blocked by the opioid antagonist naloxone. When opioids are injected into the PAG, analgesia is produced via the descending pathway.

c) Antidepressants are believed to enhance the analgesic activity of the descending pathway by increasing the availability of synaptic monoamines. The monoamines **serotonin** and **norepinephrine** are the primary neurotransmitters released by descending pathway neuron terminals. Descending pathway neurons arise in the brainstem and terminate in close proximity to primary afferent terminals, interneurons, and the synaptic membrane of second order neurons located in the dorsal horn.

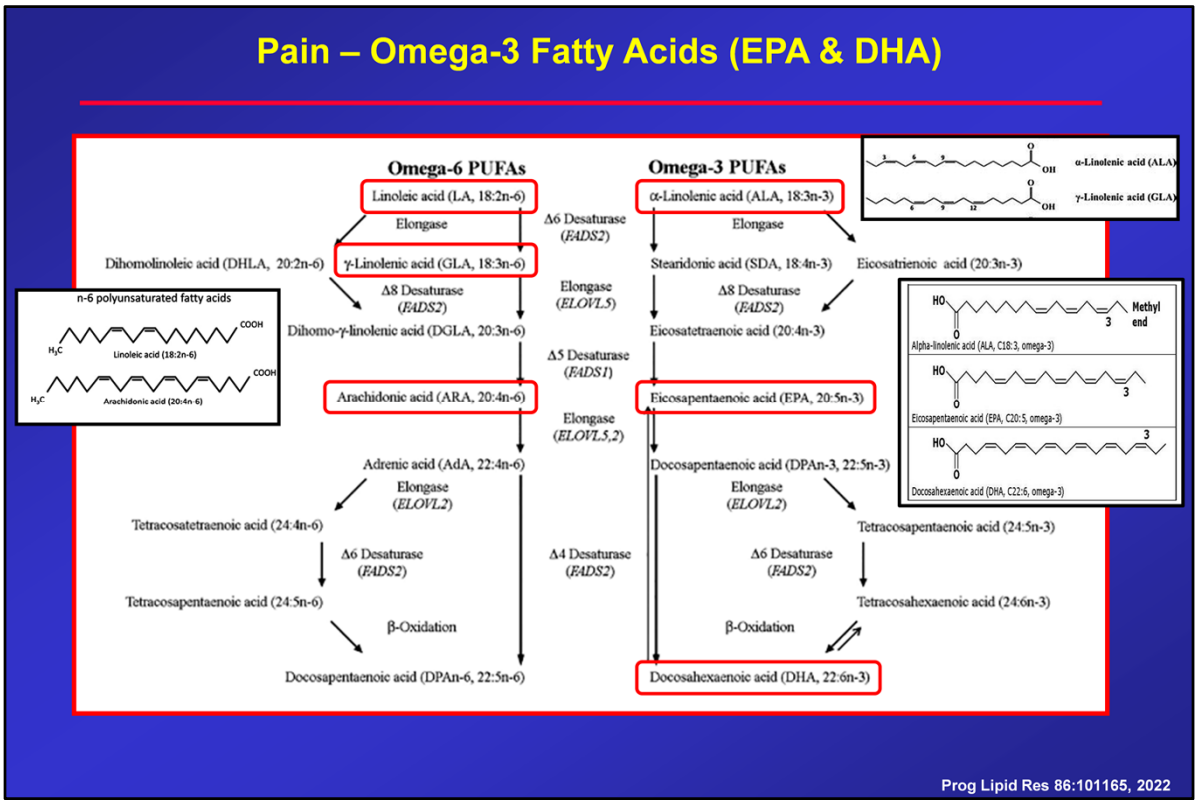
Stimulation of the nucleus raphe magnus in the brainstem results in anti-nociception

attributed to the release of **serotonin** (5-HT) within the dorsal horn. Agents that block 5-HT synthesis attenuate stimulation-produced analgesia, and the application of some 5-HT agonists in the spinal cord results in inhibition of cells responsive to nociceptive stimuli. Selective **serotonin-reuptake inhibitors** can contribute to the modulation of nociception.

Stimulation of the locus coeruleus in the medulla results in anti-nociception attributed to the release of norepinephrine within the dorsal horn. Presynaptically **noradrenaline** increases inhibitory transmitters from interneurons and depresses **glutamate** release from both Aδ and C afferent terminals.

Perception of nociceptive pain is dependent upon neural processing in the spinal cord and several brain regions. Pain becomes more than a pattern of nociceptive action potentials when they reach the brain. Action potentials ascending the **spinothalamic tract** are decoded by the thalamus, sensorimotor cortex, insular cortex, and the anterior cingulate to be perceived as an unpleasant sensation that can be localized to a specific region of the body. Action potentials ascending the **spino-bulbar tract** are decoded by the amygdala and hypothalamus to generate a sense of urgency and intensity. It is the integration of sensations, emotions and cognition that result in our perception of pain.

Positron Emission Tomography (PET) and Functional Magnetic Resonance Imaging (fMRI) have enable researchers to monitor perfusion, metabolism and the sequence of activity across multiple brain structures in response to painful stimulation. These tests demonstrate significant variation in the pattern, intensity and volume of brain activity between individuals exposed to similar painful stimulation. The findings reinforce the notion that pain is a complex individual experience that can not be quantified by anyone other than the person experiencing it.



Polyunsaturated fatty acids and fatty acid-derived lipid mediators: Recent advances in the understanding of their biosynthesis, structures, and functions
Prog Lipid Res 86:101165, 2022

Polyunsaturated fatty acids (PUFAs) are structural components of membrane phospholipids, and influence cellular function via effects on membrane properties, and also by acting as a precursor pool for lipid mediators. These lipid mediators are formed via activation of pathways involving at least one step of dioxygen-dependent oxidation, and are consequently called oxylipins. Their biosynthesis can be either enzymatically-dependent, utilizing the promiscuous cyclooxygenase, lipoxygenase, or cytochrome P450 mixed function oxidase pathways, or nonenzymatic via free radical-catalyzed pathways. The oxylipins include the classical eicosanoids, comprising prostaglandins, thromboxanes, and leukotrienes, and also more recently identified lipid mediators. With the advent of new technologies there is growing interest in identifying these different lipid mediators and characterising their roles in health and disease. This review brings together contributions from some of those at the forefront of research into lipid mediators, who provide brief introductions and summaries of current understanding of the structure and functions of the main classes of nonclassical oxylipins. The topics covered include omega-3 and omega-6 PUFA biosynthesis pathways, focusing on the roles of the different fatty acid desaturase enzymes, oxidized linoleic acid metabolites, omega-3 PUFA-derived specialized pro-

resolving mediators, elovanoids, nonenzymatically oxidized PUFAs, and fatty acid esters of hydroxy fatty acids.

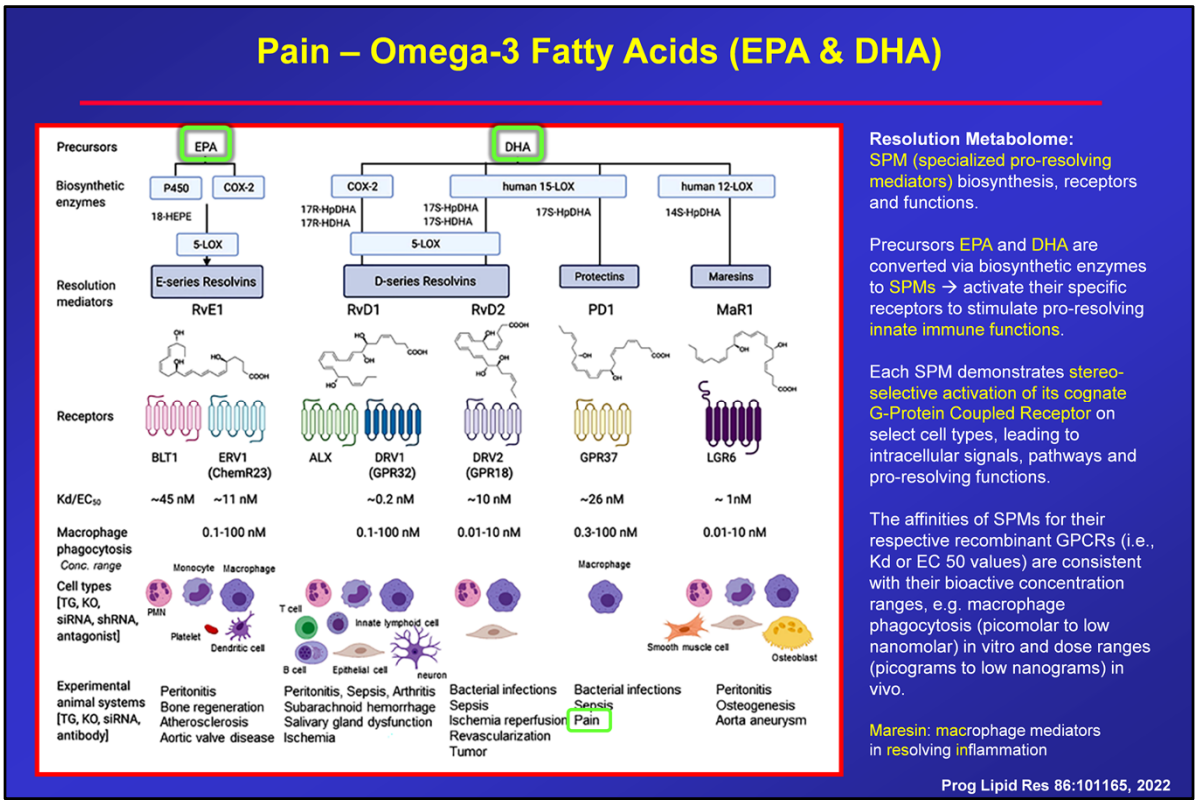


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Fig. 2.

Illustration of resolution metabolome: SPM biosynthesis, receptors and functions. Precursors EPA and DHA are converted via biosynthetic enzymes to SPMs, which in turn activate their specific receptors to stimulate pro-resolving innate immune functions. Each SPM demonstrates stereo-selective activation of its cognate GPCR on select cell types, leading to intracellular signals, pathways and pro-resolving functions. The affinities of SPMs for their respective recombinant GPCRs (i.e., Kd or EC 50 values) are consistent with their bioactive concentration ranges, e.g. macrophage phagocytosis (picomolar to low nanomolar) in vitro and dose ranges (picograms to low nanograms) in vivo. The in vivo functions of these SPM receptors were demonstrated using transgenic and/or knock-out mice, as well as specific blockage of the receptor, e.g., siRNA, antibodies or receptor antagonists (see text and recent reviews [98,116] for details).

4.2.3. SPM functions and receptors—Each SPM demonstrates potent stereo-selective actions (pico- to low nanomolar concentrations) via activation of specific G protein-coupled receptors (GPCR) on phagocytes and additional select cell types (Fig. 2).

Resolvins: Resolution phase interaction products.

E-series resolvins: RvE1 was the first identified pro-resolving molecule derived from EPA[91]. RvE1 via its receptor ERV1/ChemR23 (Kd ~11 nM) stimulates intracellular signals such as phosphorylation of S6 kinase (0.1–100 nM) (Fig. 2; reviewed in [116]).

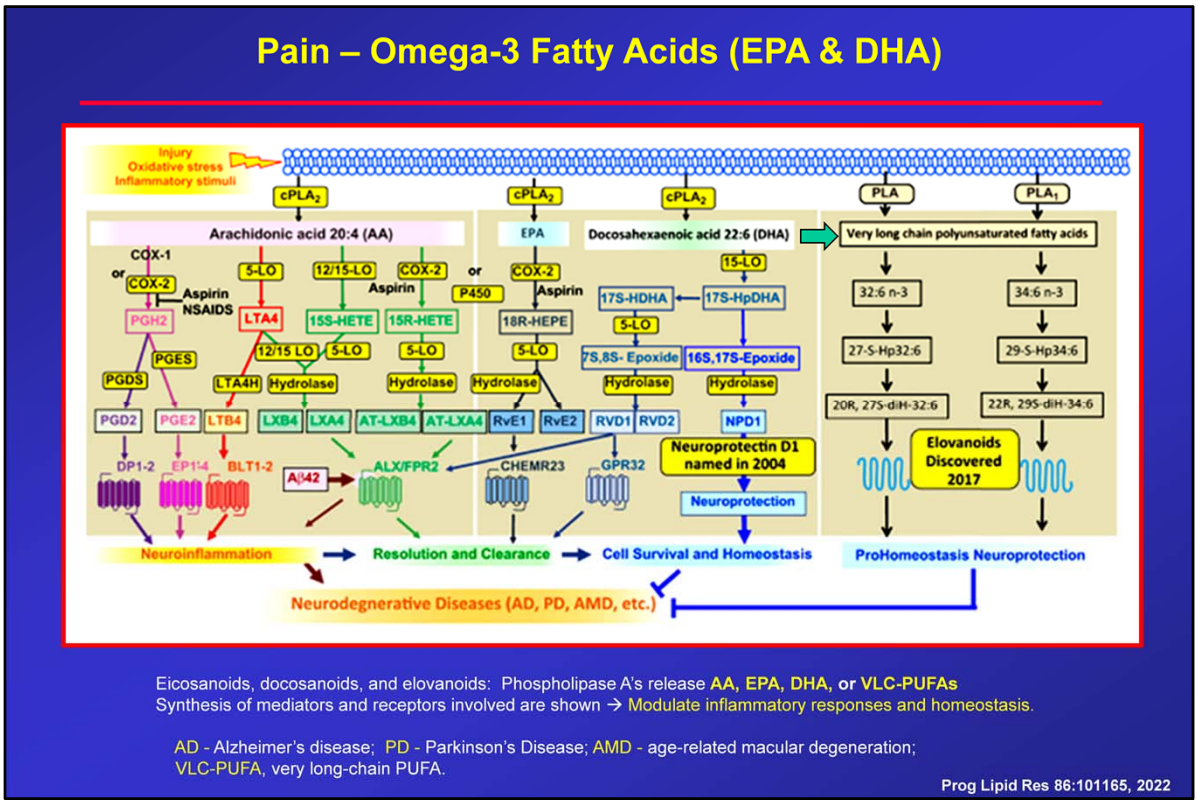
RvD1, in vivo, controls vascular inflammation, protecting against atherosclerosis by modifying oxidized LDL uptake and enhancing macrophage phagocytosis [117]. In aortic valve stenosis, targeted deletion of ChemR23 in mice heightens disease progression [118]. Of interest, an agonist antibody to the RvE1 receptor confirms that activation of the endogenous resolution mechanisms can control both inflammation and cancer burdens in mouse models in vivo [119].

D-series resolvins: RvDs are biosynthesized from DHA; they are potent immuno-resolvents active in the picomolar to low nanomolar concentrations [93,94]. RvD1 binds and activates human DRV1/GPR32 (Kd ~0.2 nM) to stimulate macrophage phagocytosis and efferocytosis (0.1–100 nM) (Fig. 2; reviewed in [101]). Some of the most exciting and unexpected findings at the time were the novel actions of RvD2. RvD2 (0.01–10 ng/mouse) limits PMN infiltration in acute inflammation and controls bacterial sepsis via its receptor DRV2/GPR18 in mice (Fig. 2, and review [98]). RvD2 binds and activates human recombinant receptor DRV2/GPR18 (Kd ~10 nM) to stimulate macrophage phagocytosis and efferocytosis (0.01–10 nM). In human sepsis, survivors had a higher percentage of GPR18-positive peripheral blood neutrophils compared to non-survivors, suggesting that DRV2/GPR18 expression levels are associated with disease severity [120]. In a more recent study, both DRV1 and DRV2 receptor expression were found to be higher on leukocytes from septic patients; both RvD1 and RvD2 partially reverse sepsis-induced leukocyte activation, and stimulate phagolysosome formation [121]. RvD2 suppresses tumor growth and enhances clearance of tumor cell debris, while DRV2/GPR18-deficient mice display defective tumor clearance [122]. In addition, RvDs are tissue/organ protective; RvD2 promotes keratinocyte repair in DRV2-dependent manner [123] and stimulates muscle regeneration [124], as well as limits tissue necrosis in burn wound [125]. RvD4 reduces thrombus burden and decreases the release of neutrophil extracellular traps (NETs), i.e. NETosis, a critical component for thrombosis development [126]. These new roles of selective RvDs suggest that SPMs could provide an effective strategy in controlling thrombo-inflammatory disease. RvD5 and RvD1 controls E. coli and S aureus infection, by controlling phagocytosis and bacterial killing as well as inflammation arising from collateral tissue damage; together these lower the antibiotic requirements for bacterial clearance [127]. Of interest, RvD5 is the first SPM that shows sex dimorphism in pain regulation, **inhibiting pain in male, but not female mice** [128].

Protectins: Protectin D1/Neuroprotectin D1 (PD1/NPD1) is also biosynthesized from DHA via 15-LOX-initiated mechanism in several human cell types, murine exudates, and brain tissues [94]. In addition, PD1 is present in human exhaled breath condensates, and its levels are lower in subjects with asthma exacerbations [129]. DHA is converted via 15-LOX to the 16S, 17S-epoxide intermediate, confirmed by epoxide trapping experiments. This epoxide intermediate is further converted to PD1 via enzymatic hydrolysis [95]. The elicited bioactivity of this mediator in human retinal pigment epithelial cells led to coining its name as Neuroprotectin D1 (NPD1) [130]. This was strongly supported by the demonstration of its formation in the human brain and its **selective decrease in memory areas of the brains of Alzheimer's patients** and in experimental Alzheimer's disease models [131,132], as well as in experimental ischemic stroke [133]. The complete stereochemical assignments [95]

enabled the demonstration of its potent actions on human PMN [1–100 nM] and acute inflammation in vivo [0.01–100 ng/mouse] as well as in many disease systems, confirmed and extended by many other investigators worldwide. Hence, while produced and functions in neural systems, the prefix (neuro)protectin D1 was introduced [130], and in the immune system, it is PD1 [134]. PD1/NPD1 displays potent neuroprotective actions in brain, retina and central nervous system, e.g. protecting from ischemic stroke, retina degenerative disease (for a recent review, see [99]) and traumatic brain injury [135]. NPD1/PD1 activates recombinant and macrophage GPR37 [EC50 ~ 26 nM]. Mice lacking this NPD1/PD1 receptor display defects in macrophage phagocytic activity with delayed resolution of inflammatory pain [136]. PD1's protective actions in multiple models of infections and sepsis are diminished in these Gpr37 receptor KO mice [137]. PDX is a positional isomer of PD1, biosynthesized via two sequential lipoxygenations [95]. PDx [0.1–10 μM] inhibits platelet activation [138], improves insulin sensitivity [139] and atherosclerosis [140] in type-2 diabetes. Both PDx and PD1 at equal amount suppress replication of influenza virus [141,142] (Fig. 3). A receptor for PDx remains to be identified. It is likely that PD1 and PDx have some overlapping yet distinct actions on select target cells.

Maresins: The macrophage mediators in resolving inflammation. MaR1 was first identifiedin self-resolving inflammatory exudates and with human macrophages (MΦ) [97] via 12-LOX-initiated mechanisms [143]. The complete stereochemistry of MaR1 was established, its total organic synthesis was achieved and confirmed by several independent teams (reviewed in [99]). MaR1 is pro-regenerative, pro-repair and neuroprotective in a wide range of tissues and organs across phyla (reviewed in [116]). MaR1 activates LGR6 (leucine-rich repeat-containing G protein–coupled receptor 6), a cell surface G protein-coupled receptor [EC50 ~ 1 nM] and stimulates key pro-resolving functions of phagocytes in a LGR6-dependent manner [0.01–10 nM] [144]. In addition, MaR1 inhibit smooth muscle cell activation and attenuate murine abdominal aortic aneurysm via LGR6 signaling [145]. Further, LGR6 is necessary for normal osteogenesis, demonstrated using LGR6-deficient mice, and MaR1 activates LGR6 signaling in osteoblasts [146]. With liver macrophages, MaR1 can also activate ROR-α (retinoic acid-related orphan receptor α), a nuclear receptor that might be relevant in liver pathology [147]. These findings highlight the cell-type specific and receptor-dependent actions of MaR1.



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4.4. Elovanoids

In the following section N.G. Bazan details the discovery of the elovanoids (ELVs) and identification of their mechanisms of action.

The significance of polyunsaturated fatty acids (PUFAs) has evolved from the broad concepts of providing membrane structural plasticity and fluidity for proteins diffusion and rotation to a diverse universe of functions. For example, DHA is necessary for sight, and when administered, is beneficial in x-linked retinitis pigmentosa and other neurode-generative diseases [178]. DHA from the diet is packaged by the liver and targeted to the central nervous system (CNS), where it achieves the highest concentration in photoreceptors and synaptic membranes [179].

PUFAs, precursors of lipid mediators and components of membrane lipids, comprise a new multidisciplinary field at the boundary of biophysics, chemical biology, and molecular physiology. Thus, at least two important issues have emerged: the gene that encodes the enzyme that elongates PUFAs to chain length ≥ 28 carbons (ELOVL4) is critically important for cell function, and

their products are precursors of the new family of lipid mediators, the elovanoids (ELVs).

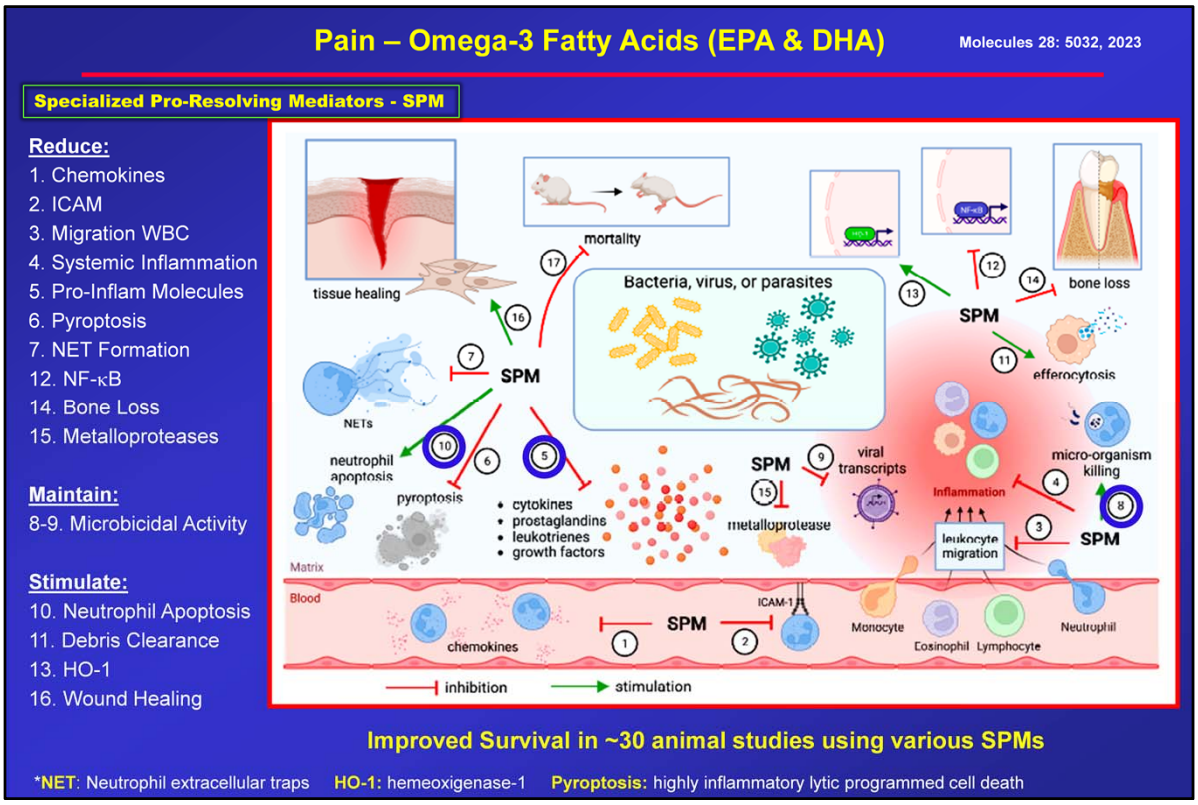
4.4.1. ELOVL4—ELOVL4 catalyzes the rate-limiting condensation reaction for the synthesis of very long chain -saturated fatty acids (VLC-SFAs) and VLCPUFAs (chain length ≥ 28 carbons) [180]. This enzyme is expressed in brain neurons, photoreceptor cells, skin, testes, and meibomian glands [180]. In the skin, VLC-SFAs are components of sphingolipids, and these VLC-SFAs are necessary as a skin-permeability barrier [181]. ELOVL4 is selectively expressed in neurons and is evolutionarily conserved [182]. In photoreceptor cells, VLC-PUFAs are in phosphatidylcholines (PC) of the outer segment membranes, tightly bound to rhodopsin [183].

Mutation, loss, or downregulation of ELOVL4 is linked to retinal degeneration. Studies of a large familial group with retinal degeneration revealed an autosomal dominant macular dystrophy phenotype which results from a 5-bp deletion, causing Stargardt-like macular dystrophy [184,185], and an STGD3 mouse Elovl4 mutation produces a C32-C36 PC deficiency [186], leading to the suggestion that loss or reduced VLC-PUFAs may cause loss of photoreceptors or functional perturbations [187], highlighting the importance of these molecules in the retina. Therefore, because of the inability to take up and incorporate DHA and the absence of VLC-PUFAs in the degenerating adiponectin receptor 1 (AdipoR1)−/− mouse retina, the synthesis of these molecules must rely on the presence of DHA. The occurrence of central geographic atrophy (CGA) and neovascular age-related macular degeneration (AMD) was found to be 30% less likely with high omega-3 LC-PUFA (e.g., DHA) intake [188], emphasizing the importance of maintaining adequate dietary amounts of DHA for retinal homeostasis.

Neuron-specific ELOVL4 is expressed in the CNS, including in hippocampal neurons of the dentate gyrus (DG) subgranular layer, a locus for medial temporal lobe epilepsy. Mutations in ELOVL4 lead to impaired neural development, mental retardation, neuronal dysfunction, hyperexcitability, and seizures [189].

4.4.2. Elovanoids are a new class of bioactive lipids synthesized from C32 or C34 FA precursors—In 2017, elovanoids (ELVs) were discovered and named [190–192]. This new class of endogenous lipid mediators is distinct from the widely known lipid mediators produced from PUFAs with C20 and C22, such as

the classical eicosanoids and SPMs. ELV-N32 and ELV-N34 are stereo-specific di-hydroxylated derivatives of 32:6n-3 or 34:6n-3 (Fig. 5), respectively, made by the elongase ELOVL4 (elongation of VLC-FAs-4), which converts C26-derived FAs from EPA or DHA to VLC-PUFAs, ≥C28. These PUFAs are mainly esterified at the C1 (sn-1) position of PC that has DHA in the C2 (sn-2) position, and upon the appropriate stimulus (e. g., uncompensated oxidative stress), are released by phospholipase A1 (PLA1) and/or PLA2 for the formation of ELVs, NPD1, or other docosanoids (Fig. 6). Here, I describe key events in the discovery of ELVs and highlight some of their functions.



Specialized Pro-Resolving Lipid Mediators: Endogenous Roles and Pharmacological Activities in Infections
Molecules 28: 5032, 2023. https://doi.org/10.3390/molecules28135032

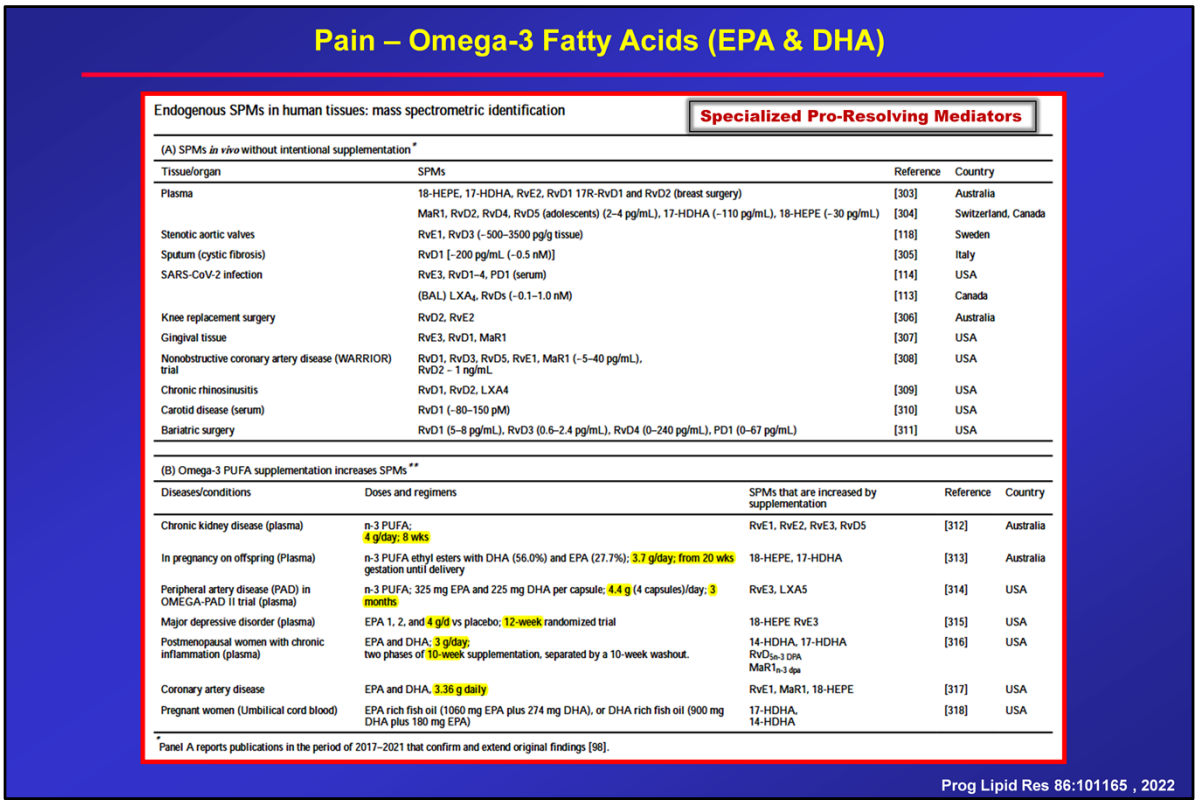
Figure 2. SPMs control infection by different mechanisms. SPMs limit chemokine release (1) and ICAM-1 expression (2), reducing migration of leukocytes (3) to the inflammatory site. Systemic inflammation (4) is also diminished, together with prominent reduction in pro-inflammatory molecules' release (5). SPMs also decrease pyroptosis (6) and NET formation (7), without compromising microbicidal activities (8 and 9). Neutrophil accumulation is avoided also by stimulating neutrophil apoptosis (10) and clearance of cell debris by macrophages (11). Inhibition of NF-B (12) and upregulation of HO-1 (13) favor inflammation control. As a result, less bone loss is observed (14), and tissue architecture is preserved by reducing the activity of metalloproteases (15) and stimulating tissue healing (16). Improvement of survival was also observed.

Abstract: During an infection, inflammation mobilizes immune cells to eliminate the pathogen and protect the host. However, inflammation can be detrimental when exacerbated and/or chronic. The resolution phase of the inflammatory process is actively orchestrated by the specialized pro-resolving lipid mediators (SPMs), generated from omega-3 and -6 polyunsaturated fatty acids (PUFAs) that bind to

different G-protein coupled receptors to exert their activity. As immuno-resolvents, SPMs regulate the influx of leukocytes to the inflammatory site, reduce cytokine and chemokine levels, promote bacterial clearance, inhibit the export of viral transcripts, enhance efferocytosis, stimulate tissue healing, and lower antibiotic requirements. Metabolomic studies have evaluated SPM levels in patients and animals during infection, and temporal regulation of SPMs seems to be essential to properly coordinate a response against the microorganism. In this review, we summarize the current knowledge on SPM biosynthesis and classifications, endogenous production profiles and their effects in animal models of bacterial, viral and parasitic infections.

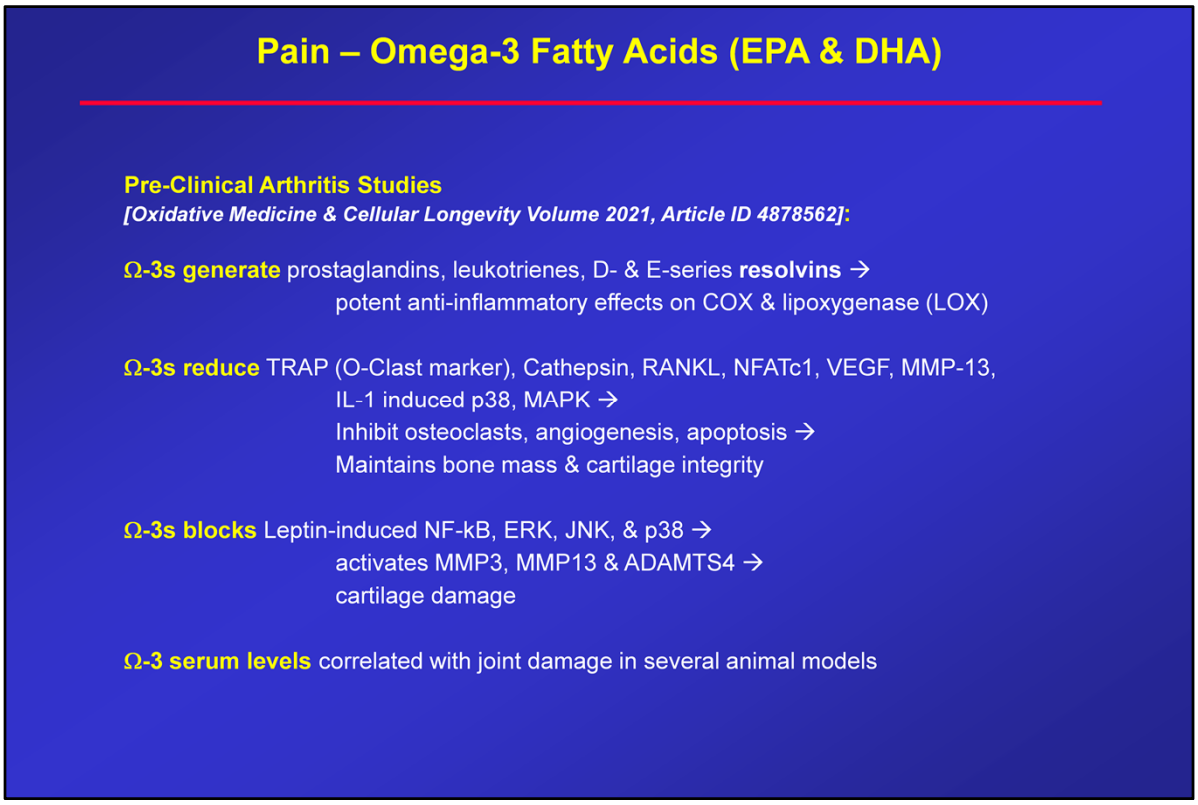
Pyroptosis is a highly inflammatory form of lytic programmed cell death

Hemeoxygenase 1 (**HO-1**) is an inducible enzyme responsible for the breakdown of heme but also exhibits numerous anti-inflammatory properties.



Prog Lipid Res 86:101165 , 2022

4.2.2. SPMs in human tissues and dysregulation in diseases—Mass spectrometry-based profiling approaches for the resolution metabolome have documented the temporal production of SPMs in humans (Table 1A) and preclinical animal systems, demonstrating in vivo the lipid mediator class switch. For example, human vagus nerves produce SPMs, e.g., RvE1, NPD1/PD1, MaR1, upon electrical stimulation [108] suggesting that this vagus-SPM circuits contribute to a new proresolving vagal reflex. Several clinical trials demonstrate omega-3 PUFA or marine oil supplementation increase SPM in vivo [109] (Table 1B). SPM biosynthesis is impaired in several diseases, including tuberculous meningitis [110], multiple sclerosis [111], and osteoarthritis [112], as well as in bronchoalveolar lavages [113], serum [114] and plasma [115] from COVID-19 patients. Thus, impaired endogenous resolution pathways may contribute to the pathogenesis of these diseases.

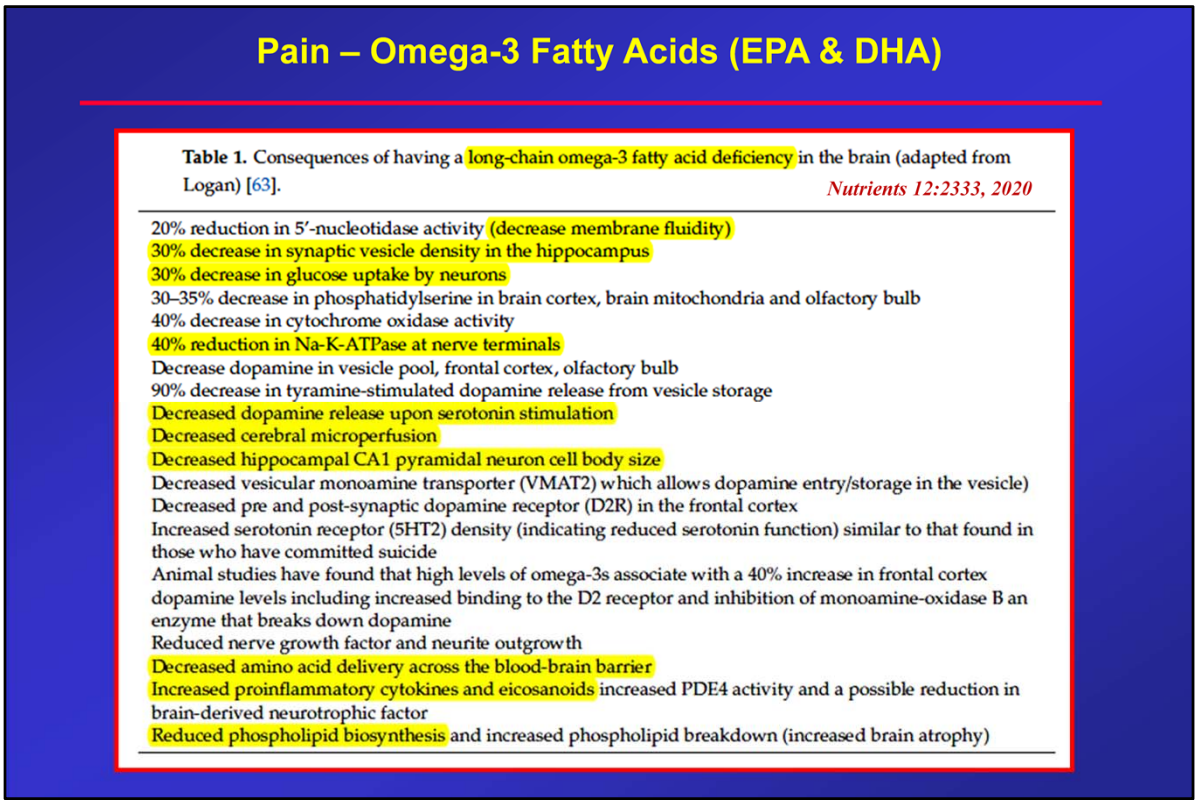


The Role of Nutraceuticals in Osteoarthritis Prevention and Treatment: Focus on n-3 PUFAs

Oxidative Medicine and Cellular Longevity Volume 2021, Article ID 4878562

Osteoarthritis (OA) is a disease caused by joint degeneration with massive cartilage loss, and obesity is among the risk factors for its onset, though the pathophysiological mechanisms underlying the disease and better therapeutic approach still remain to be assessed. In recent years, several nutraceutical interventions have been investigated in order to define better solutions for preventing and treating OA. Among them, polyunsaturated fatty acids (n-3 PUFAs) appear to represent potential candidates in counteracting OA and its consequences, due to their anti-inflammatory, antioxidant, and chondro-inductive effects. PUFAs have been found to counteract the onset and progression of OA by reducing bone and cartilage destruction, inhibiting proinflammatory cytokine release, reactive oxygen species (ROS) generation, and the NF-κB pathway's activation. Moreover, a diet rich in n-3 PUFAs and their derivatives (maresins and resolvins) demonstrates beneficial effects on associated pain reduction. Finally, it has been shown that together with the anti-inflammatory and antioxidant properties of eicosapentaenoic (EPA) and docosahexaenoic (DHA) acids, their antiapoptotic and antiangiogenic effects contribute in reducing OA development. The present review is aimed at assessing evidence suggesting the potential benefit of nutraceutical supplementation with

PUFAs in OA management according to their efficacy in targeting relevant pathophysiological mechanisms responsible for inflammation and joint destruction processes, and this may represent a novel and potentially useful approach in OA prevention and treatment. For that purpose, a PubMed literature survey was conducted with a focus on some in vitro and in vivo studies and clinical trials from 2015 to 2020.



Abstract: *Nutrients 12:2333, 2020*

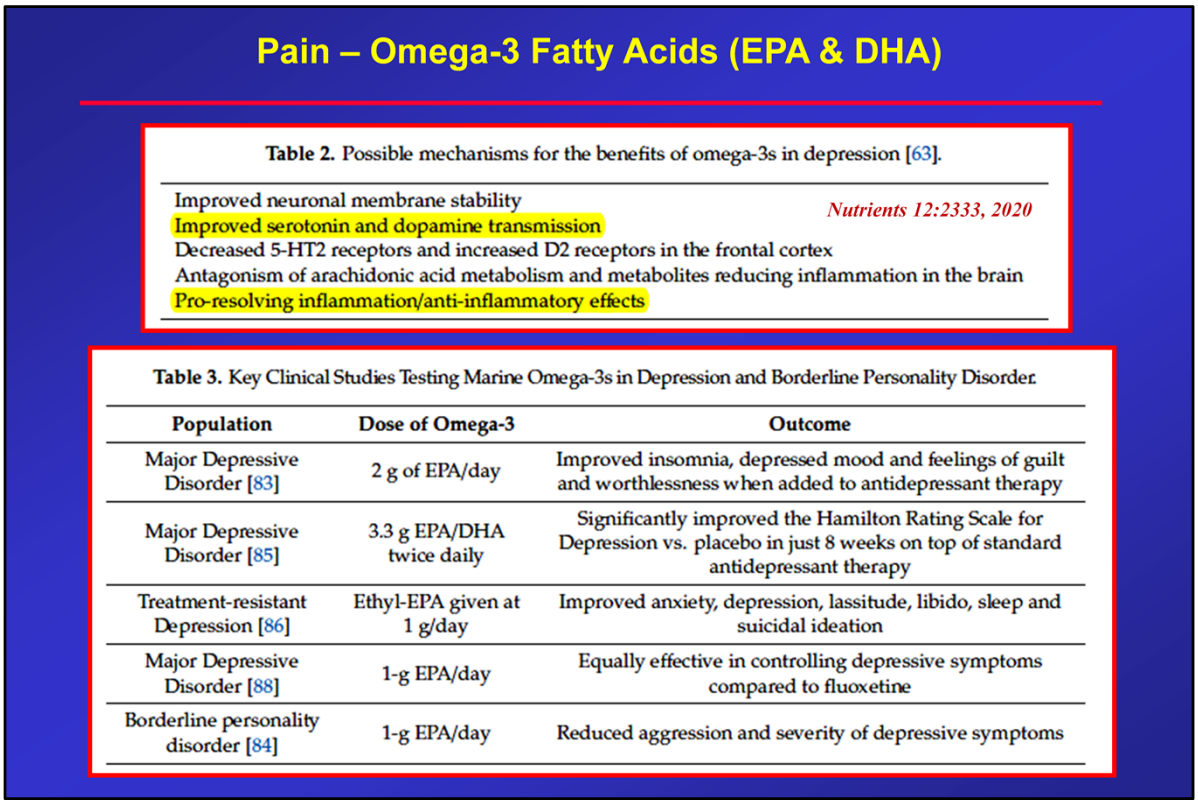
Most of the global population is deficient in long-chain marine omega-3s. In particular, docosahexaenoic acid (DHA), a long-chain omega-3 fatty acid, is important for brain and eye development. Additionally, DHA plays a significant role in mental health throughout early childhood and even into adulthood. In the brain, DHA is important for cellular membrane fluidity, function and neurotransmitter release. Evidence indicates that a low intake of marine omega-3s increases the risk for numerous mental health issues, including Attention Deficit Hyperactivity Disorder (ADHD), autism, bipolar disorder, depression and suicidal ideation. Studies giving supplemental marine omega-3s have shown promise for improving numerous mental health conditions. This paper will review the evidence surrounding marine omega-3s and mental health conditions.

9. The Importance of Long-Chain Omega-3s and Brain Health

Approximately 20% of the dry weight of the brain is made up of polyunsaturated fatty acids and one out of every three fatty acids in the nervous system is a polyunsaturated fatty acid [63]. Docosahexaenoic acid (DHA) is particularly prevalent in the brain and can be retro-converted to eicosapentaenoic acid (EPA) serving as a generator of EPA. Thus, omega-3 polyunsaturated fatty acids are extremely important in brain function and can contribute to disorders of the brain including depression. Both EPA and DHA have been shown to be important in the

treatment and prevention of depression, whereas only the latter is a major structural component of neuronal cell membrane phospholipids.

Marine omega-3s can improve neurotransmitter binding and signaling in the brain by maintaining an optimal membrane fluidity optimizing protein channel function in the lipid bi-layer [63]. Furthermore, neurological and metabolic benefits of consuming marine omega-3s may occur through activating G-protein coupled receptors (GPR40 and GPR120) as well as peroxisome proliferator-activated receptors (PPARs). Indeed, activation of PPAR-alpha can increase hepatic fatty acid oxidation and reduce triglyceride synthesis, PPAR-gamma activation can improve insulin sensitivity in adipose tissue and produce anti-inflammatory effects reducing inflammation and improving insulin sensitivity. GPR120 is highly expressed on adipocytes and inflammatory macrophages and DHA and EPA can promote GPR120-mediated gene activation inhibiting activation of nuclear factor kappa B and reducing inflammation. Furthermore, DHA promotes the translocation of the glucose transporter GLUT4 in adipocytes improving glucose uptake. All of these anti-inflammatory and metabolic effects may have a role in improving brain health. A low dietary intake of marine omega-3s reduces the concentration of omega-3s in cellular membranes, stiffening the membrane and creating a spring-like stress on protein channels which may affect their function. Indeed, low levels of long-chain omega-3 fatty acids in cellular membranes reduces the Na-K-ATPase in nerve terminals, which consumes around half the energy of the brain allowing for nerve transmission and communication [63]. Increasing long-chain omega-3s in the brain may reduce inflammatory cytokines, which may improve neurotransmitter function. There is also a reduction in dopamine and serotonin signaling with omega-3 deficiency in the brain [63]. A deficiency of omega-3 in the brain also reduces synaptic vesicle density in terminals of the hippocampus by 30%, phosphatidylserine levels in the brain by 30–35%, glucose uptake into neurons by 30%, and tyramine-stimulated dopamine release by 90% [63]. The overall consequences of having a long-chain omega-3 deficiency in the brain are summarized in Table 1 and the possible mechanisms for the benefits of omega-3s in depression are summarized in Table 2.



10. Clinical Studies Testing Marine Omega-3s in Depression and Other Brain Disorders

In a double-blind, 4-week, parallel-group study in twenty patients with a current diagnosis of major depressive disorder, 2 g of EPA/day improved insomnia, depressed mood and feelings of guilt and worthlessness when added to antidepressant therapy [83]. These benefits were noted in just three weeks after supplementation was initiated. A double-blind placebo controlled study in patents with borderline personality disorder showed that 1 g of EPA/day reduced aggression and severity of depressive symptoms [84]. Another double-blind placebo controlled study found that a total of 6.6 g of omega-3 polyunsaturated fatty acids (providing 3.3 g EPA/DHA twice daily) on top of standard
antidepressant therapy in patients with major depressive disorder significantly improved the Hamilton Rating Scale for Depression vs. placebo in just 8 weeks [85]. In patients with treatment-resistant depression, ethyl-EPA given at 1 g/day improved anxiety, depression, lassitude, libido, sleep and suicidal ideation (Table 3 summarizes the key clinical studies) [86]. Moreover, a meta-analysis of 8 randomized controlled trials in patients with depressive symptomatology but no diagnosis of major depressive disorder and 11 randomized controlled trials in patients with major depressive disordered has confirmed that omega-3 polyunsaturated fatty acids are eective in reducing depression severity
compared to placebo [87].

Most clinical trials testing marine omega-3 polyunsaturated fatty acids have found improvements in depressive disorders compared to placebo [83,86,89–91] with some showing equivalent effectiveness compared to antidepressants such as fluoxetine [88]. Omega-3s have also been found to benefit depression in those with Parkinson's disease [92] and bipolar disorder [87].

In summary, there are numerous studies and meta-analyses supporting the use of long-chain omega-3s for the prevention and treatment of major depressive disorder and patients with depressive symptoms. Considering that marine omega-3s are safe and well tolerated supplementation with EPA/DHA could be considered in those with depression or who are at an increased risk for developing depression.

Pain – Omega-3 Fatty Acids (EPA & DHA)

Box 1. Behavior, Mood, and Other Brain Disorders that May Benefit from Supplementing with Marine Omega-3s. *Nutrients 12:2333, 2020*

- Attention deficit hyperactivity disorder (ADHD) [101]
- Autism [61]
- Depression [77,102]
- Borderline personality disorder (mood instability and impulsive aggression) [84]
- Schizophrenia [103]
- Hostility [104]
- Anxiety [105]
- Bipolar disorder [106,107]
- Seasonal affective disorder [108]
- Suicidal ideation [93,94,96]

In an eight week placebo-controlled, double-blind study, 1 g of ethyl-EPA in 30 female patients with borderline personality disorder was superior to placebo in diminishing aggression as well as the severity of depressive symptoms [84]. Omega-3 polyunsaturated fatty acids may also reduce violence and suicides [93,94]. Moreover, lower levels of EPA have been noted in those with suicide attempts [95]. In patients with recurrent self-harm, supplementing with 2.1 g of EPA/DHA per day improves depression, suicidality and daily stresses [96]. Lastly, supplementation with marine omega-3s has been found to improve depressive symptoms in menopausal women with psychological distress [97], elderly depressed women [98], elderly patients with mild cognitive impairment [99] and juvenile bipolar disorder [100]. Box 1 summarizes the benefits of marine omega-3s in behavior, mood, and other brain disorders.

Pain – Omega-3 Fatty Acids (EPA & DHA)

Diabetic Neuropathy:

21 T2DM: EPA-only 1,800 mg for 48 weeks (no controls) → Improved Coldness, & Vibration Perception (p<0.01)
Mostly within 12 weeks. *J Diab Comp 10:280-287, 1996*

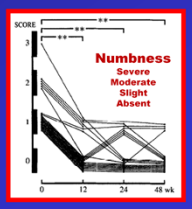


TABLE 2. CHANGES IN VIBRATORY PERCEPTION THRESHOLD (VPT) OF TIBIA AND ULNA DURING TREATMENT WITH EPA-E

	Before	During		
	0 weeks	12 weeks	24 weeks	48 weeks
VPT Tibia (μm)	32.1 ± 8.5	19.1 ± 3.6*	22.1 ± 6.8**	16.1 ± 4.8**
Ulna (μm)	13.3 ± 1.4	9.5 ± 0.4*	9.6 ± 0.7	9.8 ± 2.2**

26 T2DM: DHA 1,000 mg + EPA 200 mg for 12 weeks (no controls) → improved SF-MPQ scores:
Sensory (p<0.001), Affective (p=0.012) & Visual (p<0.001) analogue scales.
Diabetes, Metabolic Syndrome and Obesity: Targets and Therapy 2019:12

	All participants with neuropathic pain symptoms			Low presupplementation, SF-MPQ scores short-form McGill Pain Questionnaire			Moderate–high presupplementation, SF-MPQ scores		
n	26			14			12		
Age, years	52.9 (10.7)			55.2 (9.4)			50.3 (11.9)		
Male, %	34.6			28.6			41.7		
Latino, %	100								
SF-MPQ scores	**Baseline**	**3 months**	***P*-value**	**Baseline**	**3 months**	***P*-value**	**Baseline**	**3 months**	***P*-value**
Sensory	7.7 (5.7)	2.4 (2.4)	<0.001	3.4 (1.7)	1.3 (1.9)	0.014	12.8 (4.3)	3.6 (2.4)	0.002
Affective	1.9 (2.4)	0.6 (1.1)	0.012	0.9 (1.7)	0.6 (1.3)	0.596	3.1 (2.6)	0.58 (0.9)	0.017
VAS, mm	36.5 (21.2)	15.4 (18.4)	<0.001	30.7 (20.6)	12.1 (19.3)	0.016	43.3 (20.5)	19.2 (17.3)	0.007

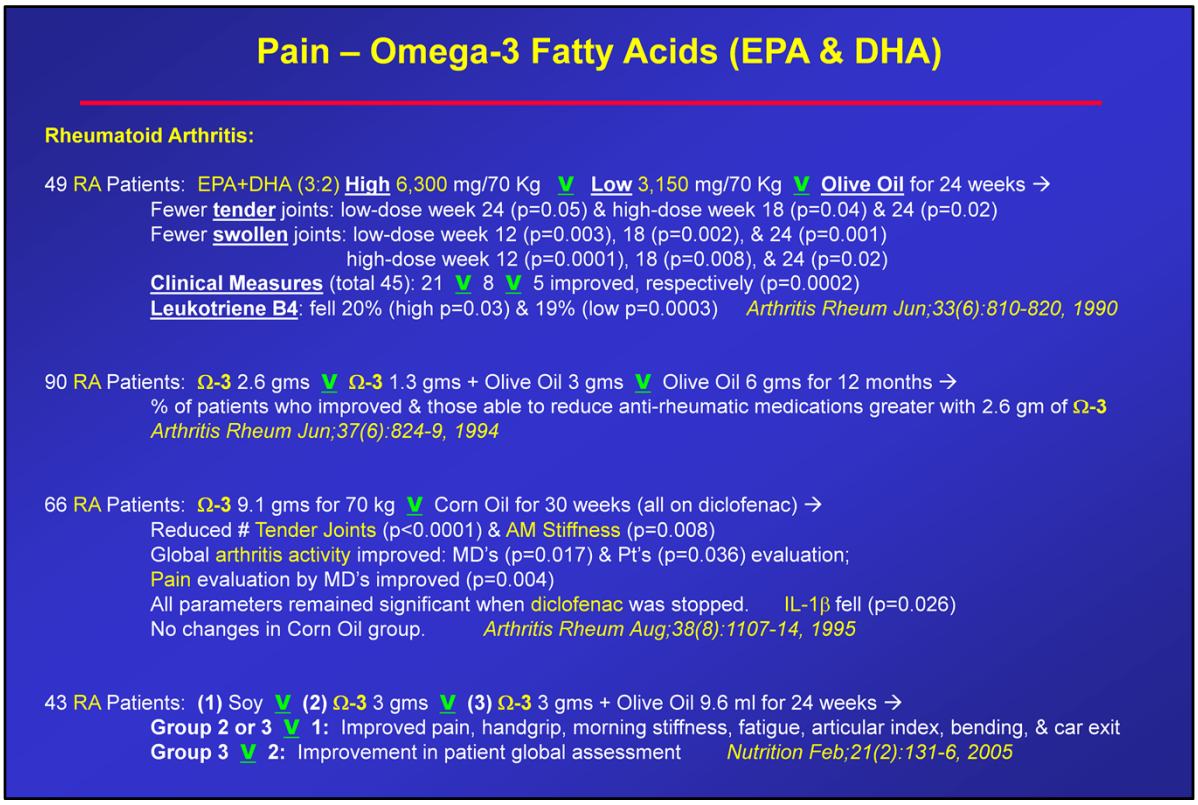
J Diab Comp 10:280-287, 1996 **ABSTRACT**

The present study was undertaken to investigate the efficacy of a new, highly purified (purity greater than 91%), ethyl esterification product from natural eicosapentaenoic acid (EPA-E, C20:5 03) in patients with type II diabetes mellitus (NIDDM). Hemodynamic changes were assessed at the level of the dorsalis pedis artery using an ultrasonic color Doppler duplex system before and after oral administration of EPA-E at a dose of **1800 mg/day** for 48 weeks. The cross-sectional **area of the dorsalis pedis artery** increased significantly from 2.5±0.2 to 3.9±0.4 mm^2 (48 weeks, mean±SE, $p < 0.05$). Moreover, EPA-E improved the clinical symptom (**coldness, numbness**) as well as the **vibration perception threshold sense** of the lower extremities [from 32.1±8.5 to 16.1±4.8 (48 weeks) micrometers]. A significant decrease of serum **triglycerides** was also noted by EPA-E administration. Furthermore, significant decrease of the excretion of albumin in urine from 24.4±3.3 to 13.9±1.8 (48 weeks) mg/gm Cr, $p<0.051$. The results of this study suggest that EPA-E has significant beneficial effects on diabetic neuropathy and serum lipids as well as other diabetic complications such as nephropathy and macroangiopathy.

Diabetes, Metabolic Syndrome and Obesity: Targets and Therapy 2019:12 **Abstract**

Purpose: To determine whether dietary supplementation with omega-3 polyunsaturated fatty acids (PUFAs) reduces neuropathic pain symptoms in Mexican-

Americans with type 2 diabetes.
Methods: Forty volunteers with type 2 diabetes enrolled in the "En Balance-PLUS" program, which provided weekly nutrition–diabetes education and daily supplementation with 1,000 mg docosahexaenoic acid (DHA) & 200 mg eicosapentaenoic acid (EPA) over 3 months. The study assessed self-reported neuropathic pain symptoms pre/postintervention using the short-form McGill Pain Questionnaire (SF-MPQ), monitored clinical laboratory values at baseline and 3 months, and performed baseline and 3-month metabolomic analysis of plasma samples.
Results: A total of 26 participants self-reported neuropathic pain symptoms at baseline. After 3 months of omega-3 PUFA supplementation, participants reported significant improvement in SF-MPQ scores (sensory, affective, and visual analogue scale; $P<0.001$, $P=0.012$, and $P<0.001$, respectively). Untargeted metabolomic analysis revealed that participants in the moderate–high SF-MPQ group had the highest relative plasma sphingosine levels at baseline compared to the low SF-MPQ group ($P=0.0127$) and the non-pain group ($P=0.0444$). Omega-3 PUFA supplementation increased plasma DHA and reduced plasma sphingosine levels in participants reporting neuropathic pain symptoms ($P<0.001$ and $P<0.001$, respectively). Increased plasma DHA levels significantly correlated with improved SF-MPQ sensory scores ($r=0.425$, $P=0.030$). Improved SF-MPQ scores, however, did not correlate with clinical/laboratory parameters.
Conclusion: The data suggest that omega-3 PUFAs dietary supplementation may reduce neuropathic pain symptoms in individuals with type 2 diabetes and correlates with sphingosine levels in the plasma.



Arthritis Rheum Jun;33(6):810-820, 1990 **Abstract**

Forty-nine patients with active rheumatoid arthritis completed a 24-week, prospective, double-blind, randomized study of dietary supplementation with 2 different dosages of fish oil and 1 dosage of olive oil. Clinical evaluations were performed at baseline and every 6 weeks thereafter, and immunologic variables were measured at baseline and after 24 weeks of study. The 3 groups of patients were matched for age, sex, disease severity, and use of disease-modifying antirheumatic drugs (DMARDs). Subjects continued receiving DMARDs and other background medications without change during the study.

Twenty patients consumed daily dietary supplements of n3 fatty acids containing 27 mg/kg eicosapentaenoic acid (EPA 1890 mg for 70 kg) and 18 mg/kg docosahexaenoic acid (DHA 1260 mg) (low dose – 3150 mg/70 kg), 17 patients ingested 54 mg/kg EPA (3780 mg) and 36 mg/kg DHA (2520 mg) (high dose – 6300 mg/70 kg), and 12 patients ingested olive oil capsules containing 6.8 gm of oleic acid.

Significant improvements from baseline in the number of tender joints were noted in the low-dose group at week 24 (P = 0.05) and in the high-dose group at week 18 (P = 0.04) and 24 (P = 0.02). Significant decreases from baseline in the number of swollen joints were noted in the low-dose group at weeks 12 (P = 0.003), 18 (P = 0.002), and 24 (P = 0.001) and in the high-dose group at weeks 12 (P = 0.0001), 18 (P = 0.008), and 24 (P = 0.02). A total of 5 of 45 clinical measures were significantly

changed from baseline in the olive oil group, 8 of 45 in the low-dose fish oil group, and 21 of 45 in the high-dose fish oil group during the study (P = 0.0002). Neutrophil leukotriene B4 production decreased by 19% from baseline in the low-dose fish oil group (P = 0.0003) and 20% in the high-dose group (P = 0.03), while macrophage interleukin-1 production decreased by 38.5% in the olive oil group (P not significant), 40.6% in the low-dose group (P = 0.06), and 54.7% in the high-dose group (P = 0.0005). Tritiated thymidine incorporation in peripheral blood mononuclear cells after stimulation with concanavalin A increased significantly in all 3 groups after 24 weeks, compared with baseline values.
We conclude that the clinical benefits of dietary supplementation with omega-3 fatty acids are more commonly observed in patients consuming higher dosages of fish oil for time intervals that are longer than those previously studied. Dietary supplementation with olive oil is also associated with certain changes in immune function, which require further investigation.

Arthritis Rheum Jun;37(6):824-9, 1994 **Abstract**

Objective: To study the long-term effects of supplementation with omega-3 fatty acids (omega 3) in patients with active rheumatoid arthritis.
Methods: Ninety patients were enrolled in a 12-month, double-blind, randomized study comparing daily supplementations with either 2.6 gm of omega 3, or 1.3 gm of omega 3 + 3 gm of olive oil, or 6 gm of olive oil.
Results: Significant improvement in the patient's global evaluation and in the physician's assessment of pain was observed only in those taking 2.6 gm/day of omega 3. The proportions of patients who improved and of those who were able to reduce their concomitant anti-rheumatic medications were significantly greater with 2.6 gm/day of omega 3.
Conclusion: Daily supplementation with 2.6 gm of omega 3 results in significant clinical benefit and may reduce the need for concomitant anti-rheumatic medication.

Arthritis Rheum Aug;38(8):1107-14, 1995 **Abstract**

Objective: To determine the following: 1) whether dietary supplementation with fish oil will allow the discontinuation of nonsteroidal anti-inflammatory drugs (NSAIDs) in patients with rheumatoid arthritis (RA); 2) the clinical efficacy of high-dose dietary omega 3 fatty acid fish oil supplementation in RA patients; and 3) the effect of fish oil supplements on the production of multiple cytokines in this population.
Methods: Sixty-six RA patients entered a double-blind, placebo-controlled, prospective study of fish oil supplementation while taking diclofenac (75 mg twice a day). Patients took either 130 mg/kg/day (9100 mg for 70 kg) of omega 3 fatty acids or 9 capsules/day of corn oil. Placebo diclofenac was substituted at week 18 or 22, and fish oil supplements were continued for 8 weeks (to week 26 or 30). Serum

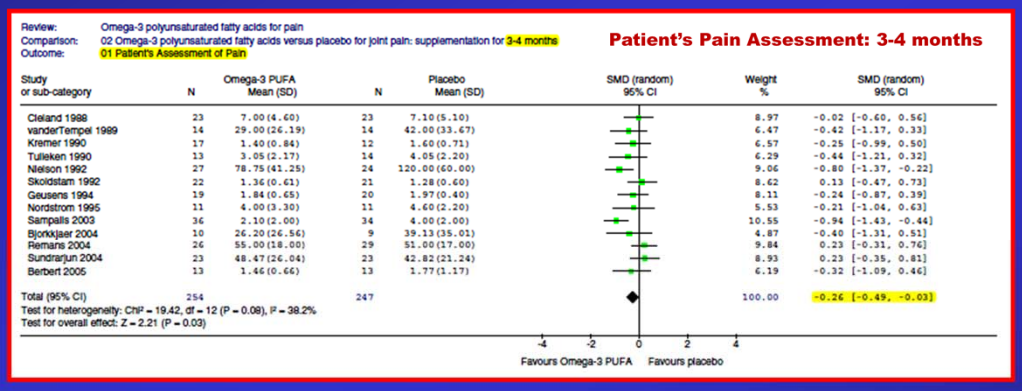
levels of interleukin-1 beta (IL-1 beta), IL-2, IL-6, and IL-8 and tumor necrosis factor alpha were measured by enzyme-linked immunosorbent assay at baseline and during the study.
Results: In the group taking fish oil, there were significant decreases from baseline in the mean (+/- SEM) number of tender joints (5.3 +/- 0.835; P < 0.0001), duration of morning stiffness (-67.7 +/- 23.3 minutes; P = 0.008), physician's and patient's evaluation of global arthritis activity (-0.33 +/- 0.13; P = 0.017 and -0.38 +/- 0.17; P = 0.036, respectively), and physician's evaluation of pain (-0.38 +/- 0.12; P = 0.004). In patients taking corn oil, no clinical parameters improved from baseline. The decrease in the number of tender joints remained significant 8 weeks after discontinuing diclofenac in patients taking fish oil (-7.8 +/- 2.6; P = 0.011) and the decrease in the number of tender joints at this time was significant compared with that in patients receiving corn oil (P = 0.043). IL-1 beta decreased significantly from baseline through weeks 18 and 22 in patients consuming fish oil (-7.7 +/- 3.1; P = 0.026).
Conclusion: Patients taking dietary supplements of fish oil exhibit improvements in clinical parameters of disease activity from baseline, including the number of tender joints, and these improvements are associated with significant decreases in levels of IL-1 beta from baseline. Some patients who take fish oil are able to discontinue NSAIDs without experiencing a disease flare.

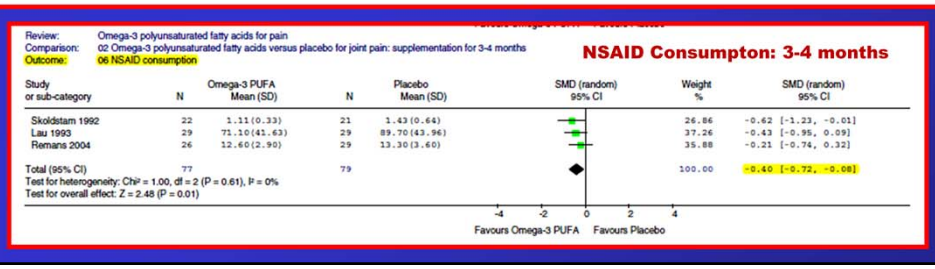
Nutrition Feb;21(2):131-6, 2005 **Abstract**
Objective: This study evaluated whether supplementation with olive oil could improve clinical and laboratory parameters of disease activity in patients who had rheumatoid arthritis and were using fish oil supplements.
Methods: Forty-three patients (34 female, 9 male; mean age = 49 +/- 19y) were investigated in a parallel randomized design. Patients were assigned to one of three groups. In addition to their usual medication, the first group (G1) received placebo (soy oil), the second group (G2) received fish oil omega-3 fatty acids (3 g/d), and the third group (G3) received fish oil omega-3 fatty acids (3 g/d) and 9.6 mL of olive oil. Disease activity was measured by clinical and laboratory indicators at the beginning of the study and after 12 and 24 wk. Patients' satisfaction in activities of daily living was also measured.
Results: There was a statistically significant improvement (P < 0.05) in G2 and G3 in relation to G1 with respect to joint pain intensity, right and left handgrip strength after 12 and 24 wk, duration of morning stiffness, onset of fatigue, Ritchie's articular index for pain joints after 24 wk, ability to bend down to pick up clothing from the floor, and getting in and out of a car after 24 wk. G3, but not G2, in relation to G1 showed additional improvements with respect to duration of morning stiffness after 12 wk, patient global assessment after 12 and 24 wk, ability to turn faucets on and off after 24 wk, and rheumatoid factor after 24 wk. In addition, G3 showed a significant

improvement in patient global assessment in relation to G2 after 12 wk.
Conclusions: Ingestion of fish oil omega-3 fatty acids relieved several clinical parameters used in the present study. However, patients showed a more precocious and accentuated improvement when fish oil supplements were used in combination with olive oil.

Pain – Omega-3 Fatty Acids (EPA & DHA)

Rheumatoid Arthritis (continued): *Pain 129:210–223, 2007* – **Meta-analysis**



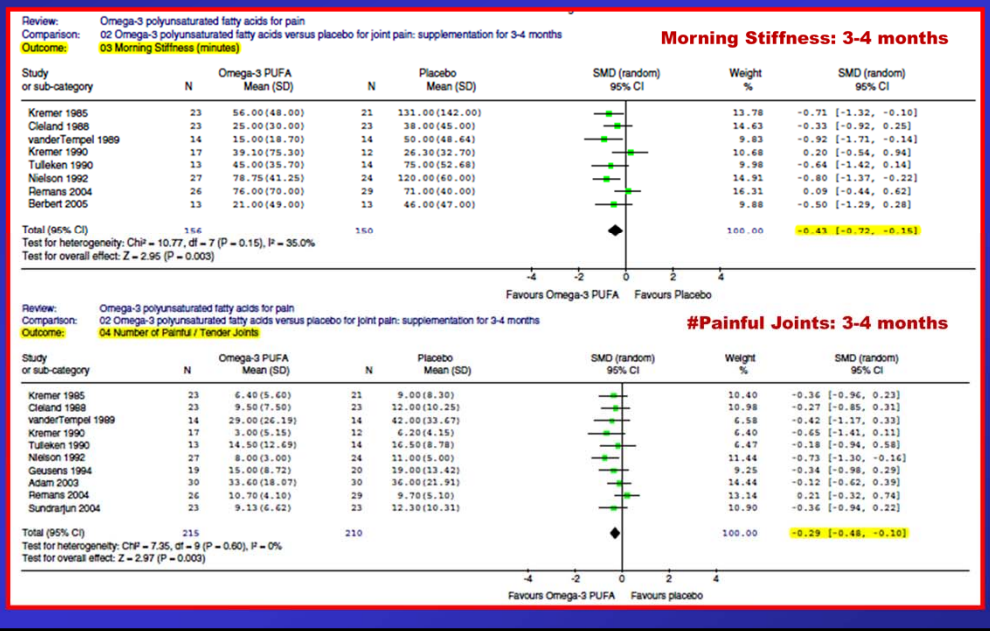


Pain 129:210–223, 2007 **Abstract**

Between 40% and 60% of Americans use complementary and alternative medicine to manage medical conditions, prevent disease, and promote health and well-being. Omega-3 polyunsaturated fatty acids (Omega-3 PUFAs) have been used to treat joint pain associated with several inflammatory conditions. We conducted a meta-analysis of 17 randomized, controlled trials assessing the pain relieving effects of omega-3 PUFAs in patients with rheumatoid arthritis or joint pain secondary to inflammatory bowel disease and dysmenorrhea. Meta-analysis was conducted with Cochrane Review Manager for six separate outcomes using standardized mean differences (SMDs) as a measure of effect size: (1) patient assessed pain, (2) physician assessed pain, (3) duration of morning stiffness, (4) number of painful and/or tender joints, (5) Ritchie articular index, and (6) nonselective nonsteroidal anti-inflammatory drug consumption. Supplementation with omega-3 PUFAs for 3–4 months reduces patient reported joint pain intensity (SMD: 0.26; 95% CI: 0.49 to 0.03, p = 0.03), minutes of morning stiffness (SMD: 0.43; 95% CI: 0.72 to 0.15, p = 0.003), number of painful and/or tender joints (SMD: 0.29; 95% CI: 0.48 to 0.10, p = 0.003), and NSAID consumption (SMD: 0.40; 95% CI: 0.72 to 0.08, p = 0.01). Significant effects were not detected for physician assessed pain (SMD: 0.14; 95% CI: 0.49 to 0.22, p = 0.45) or Ritchie articular index (SMD: 0.15; 95% CI: 0.19 to 0.49, p = 0.40) at 3–4 months.

Pain – Omega-3 Fatty Acids (EPA & DHA)

Rheumatoid Arthritis (continued): *Pain 129:210–223, 2007* – **Meta-analysis**



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Pain – Omega-3 Fatty Acids (EPA & DHA)

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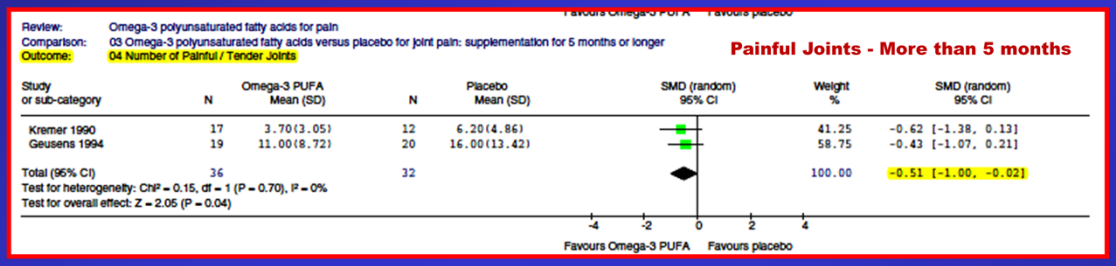

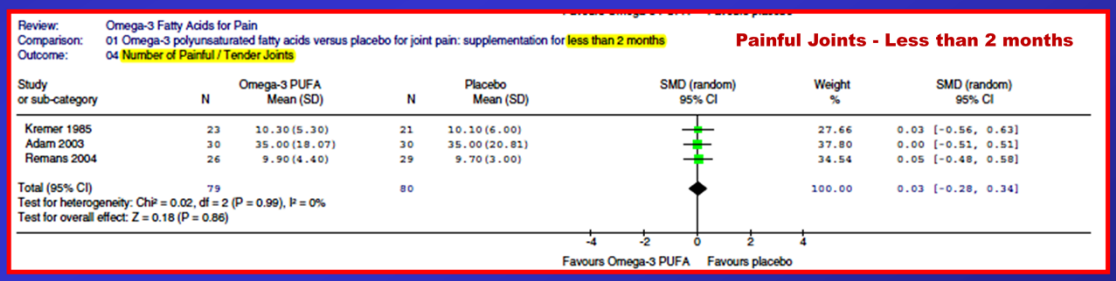

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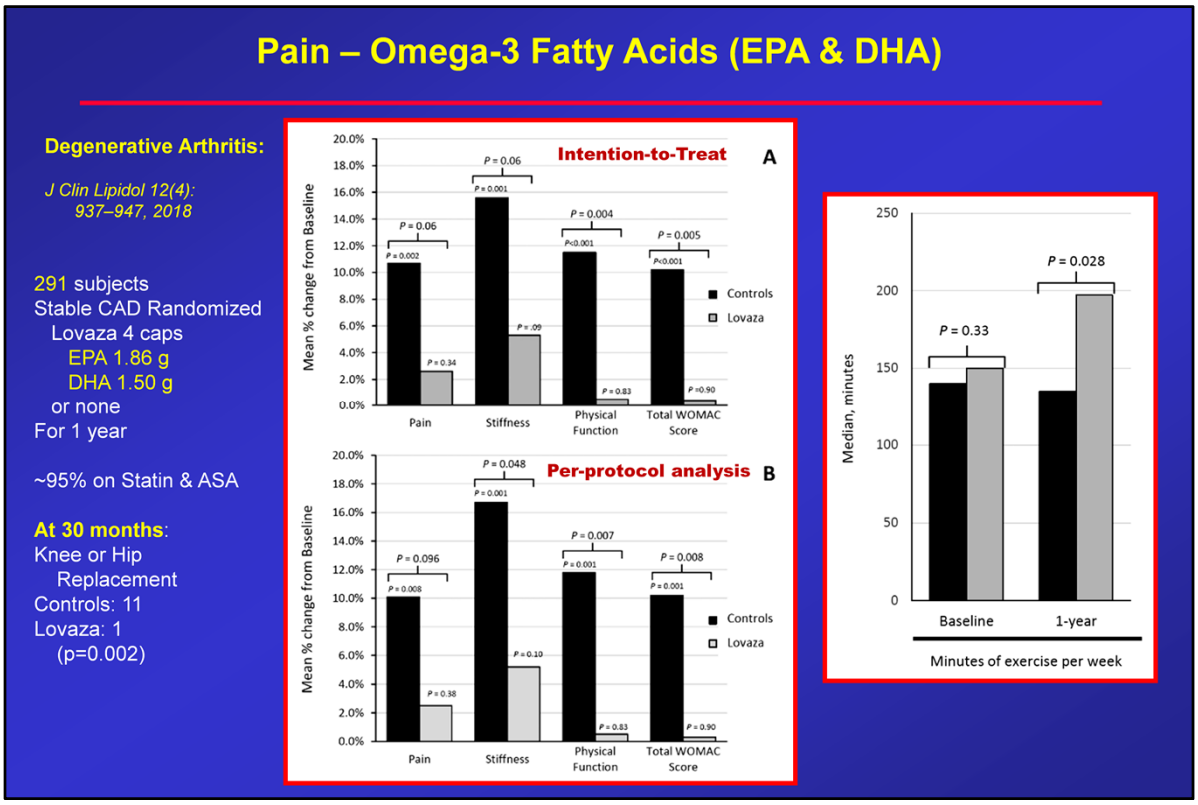


BACKGROUND: Poor physical function impairs fitness and exercise and is associated with worse cardiovascular outcomes and all-cause mortality. Joint pain and stiffness limit physical function.

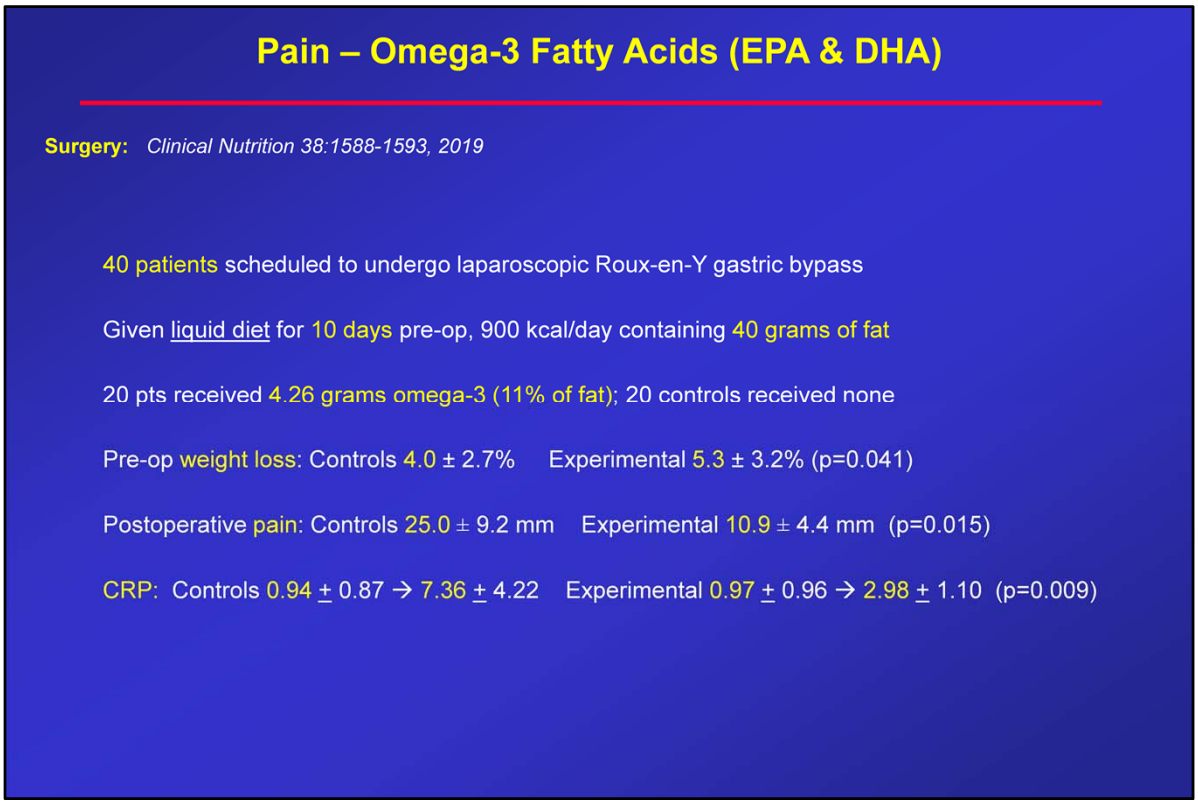
OBJECTIVE: To determine if eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) supplementation improves physical function and exercise in coronary artery disease (CAD) patients.

METHODS: 291 subjects with stable CAD were randomized to either Lovaza (1.86 g of EPA and 1.5 g of DHA daily) or no Lovaza (control) for 1 year. Change in pain, stiffness and physical function was assessed by the Western Ontario and McMaster Universities Arthritis Index (WOMAC). Minutes of exercise per week were recorded, and musculoskeletal events were reported.

RESULTS: Mean age (SD) was 63.3 (7.6) years. In the intention-to-treat analysis, compared to controls, those on Lovaza had better physical function (mean difference, −11.0%, 95% CI −18.5% to −3.5%, P = .004), better total WOMAC scores (mean difference, −9.8%, 95% CI −16.6% to −3.0%, P = .005), more exercise per week (135 minutes versus 197 minutes, respectively, P = .028) and less joint replacement

(11 vs 1, respectively, P = .002). Pain and stiffness showed a trend toward significance (P = .06). The per-protocol analysis also showed less stiffness compared to controls (mean difference, −11.5%, 95% CI −22.9% to −0.1%, P = .048).

CONCLUSION: High-dose EPA and DHA may benefit CAD patients by preserving physical function, increasing amount of exercise and reducing joint replacement. EPA and DHA may be a safe preventative strategy against musculoskeletal symptoms in CAD patients.



Background: The term "Immunonutrition" (IMN) describes the enteral administration of certain substrates with a theoretical immunomodulating function. From all the elements conforming these IMN formulas, Omega-3 fatty acids (O3FA) are hypothesized to be the most important component for immunomodulation, with increased anti-inflammatory and antioxidant effect.

Patients and methods: A prospective randomized clinical trial of all the patients undergoing laparoscopic Roux-en-Y gastric bypass was performed. Patients were randomly assigned into 2 groups: those patients receiving a preoperative balanced energy high-protein formula (Control Group) and those ones who received the same preoperative nutritional formula enriched with O3FA (Experimental Group). In both groups, there was a restriction to 900 Kcal/day. Nutritional intervention started 10 days before surgery and was maintained up to 8 h before the surgical act. Preoperative weight loss, postoperative pain, complications and acute phase reactants were investigated.

Results: 40 patients were included in the study, 20 in each group. Preoperative excess weight loss (EWL) with the prescribed treatment was 10.6 ± 7.7% in Control Group and 14.1 ± 5.8% in the Experimental Group (p=0.024). Mean postoperative pain was 25.0 ± 9.2 mm in Control group and 10.9 ± 4.4 mm in Experimental Group

(p=0.015). CRP determined 24 h after surgery was significantly lower in the Experimental Group than in the Control Group. There were not significant differences in complications, mortality or readmission rates between groups.

Conclusions: The use of a nutritional supplement enriched with O3FA is associated with a greater preoperative weight loss, reduced postoperative pain and decreased postoperative levels of C reactive protein.

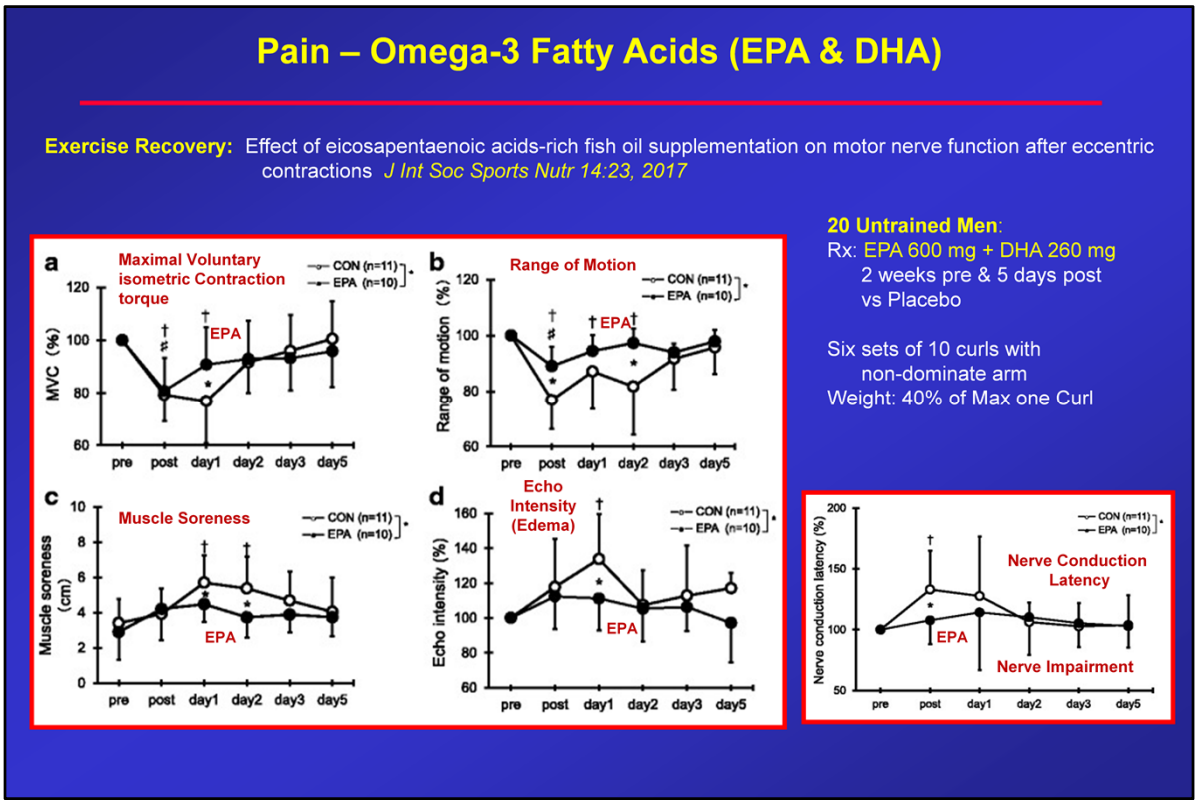


Fig. 1

Changes (mean ± SD) of maximal voluntary isometric contraction (MVC) torque (**a**), range of motion (ROM) (**b**), muscle soreness (**c**), and echo intensity (**d**) measured before (pre) and immediately after (post) the eccentric contractions exercise and then 1, 2, 3, and 5 days later in the control and EPA groups. The values of MVC torque, ROM, and echo intensity at post, 1, 2, 3, and 5 days after eccentric contractions were calculated by relative changes from baseline (100%). * $p < 0.05$ for the difference between groups; † $p < 0.05$ for the difference from the pre-exercise value in the control group, # $p < 0.05$ for the difference from pre-exercise value in the EPA group

Fig. 3

Changes (mean ± SD) of nerve conduction latency measured before (pre) and immediately after (post) the eccentric contractions exercise and then 1, 2, 3, and 5 days later in the control and EPA groups. The values of nerve conduction latency at post, 1, 2, 3, and 5 days

Nerve conduction velocity, M-wave latency, and amplitude are often measured to assess motor nerve function [6, 7]. M-wave latency is measured as the time between electrical stimulation and the onset of an M-wave, but this can be influenced by a number of factors, including various processes such as nerve conduction, neuromuscular transmission, and muscle fiber conduction, and sarcolemmal

excitability [8]. Previous studies have used M-wave latency to examine nerve disorders such as neuropathy and neural muscular atrophy [6, 9]; these indicated that an increase in M-wave latency reflected the impairment of motor nerves. It has been shown that ECCs (eccentric contractions) caused histological damage in rats, not only in myofibrils, the extracellular matrix, and the triads of the cytoplasmic membrane system [10, 11], but also in nerve fibers and in the thinning of myelin sheaths [12]. Kouzaki et al. [13] reported that M-wave latency delayed by 12% at 24 h and 24% at 48 h after 60 ECCs of the elbow flexors performed by women, suggesting musculocutaneous nerve impairment. However, it is not known whether nutritional strategies have a role for preventing temporal muscular dysfunction after ECCs.

Abstract Background

This study investigated the effect of supplementation with fish oil rich in eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) on the M-wave latency of biceps brachii and muscle damage after a single session of maximal elbow flexor eccentric contractions (ECC).

Methods

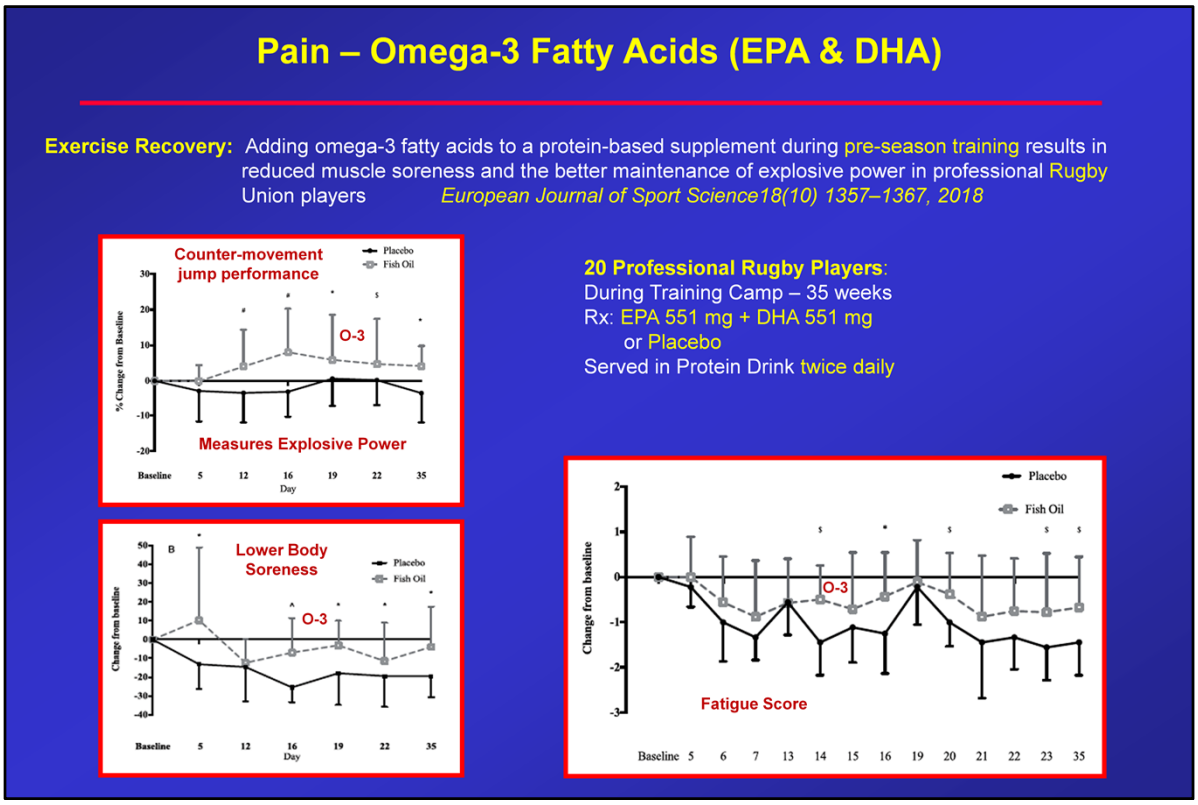
Twenty-one men were completed the randomized, double-blind, placebo-controlled, and parallel-design study. The subjects were randomly assigned to the fish oil group (n = 10) or control group (n = 11). The fish oil group consumed eight 300-mg EPA-rich fish oil softgel capsules (containing, in total, 600 mg EPA and 260 mg DHA) per day for 8 weeks *(low dose 860 mg O-3)* before the exercise, and continued this for a further 5 days. The control group consumed an equivalent number of placebo capsules. The subjects performed six sets of ten eccentric contractions of the elbow flexors using a dumbbell set at 40% of their one repetition maximum. M-wave latency was assessed as the time taken from electrical stimulation applied to Erb's point to the onset of M-wave of the biceps brachii. This was measured before and immediately after exercise, and then after 1, 2, 3, and 5 days. Changes in maximal voluntary isometric contraction (MVC) torque, range of motion (ROM), upper arm circumference, and delayed onset muscle soreness (DOMS) were assessed at the same time points.

Results

Compared with the control group, M-wave latency was significantly shorter in the fish oil group immediately after exercise (p = 0.040), MVC torque was significantly higher at 1 day after exercise (p = 0.049), ROM was significantly greater at post and 2 days after exercise (post; p = 0.006, day 2; p = 0.014), and there was significantly less delayed onset muscle soreness at 1 and 2 days after exercise (day 1; p = 0.049, day 2; p = 0.023).

Conclusion

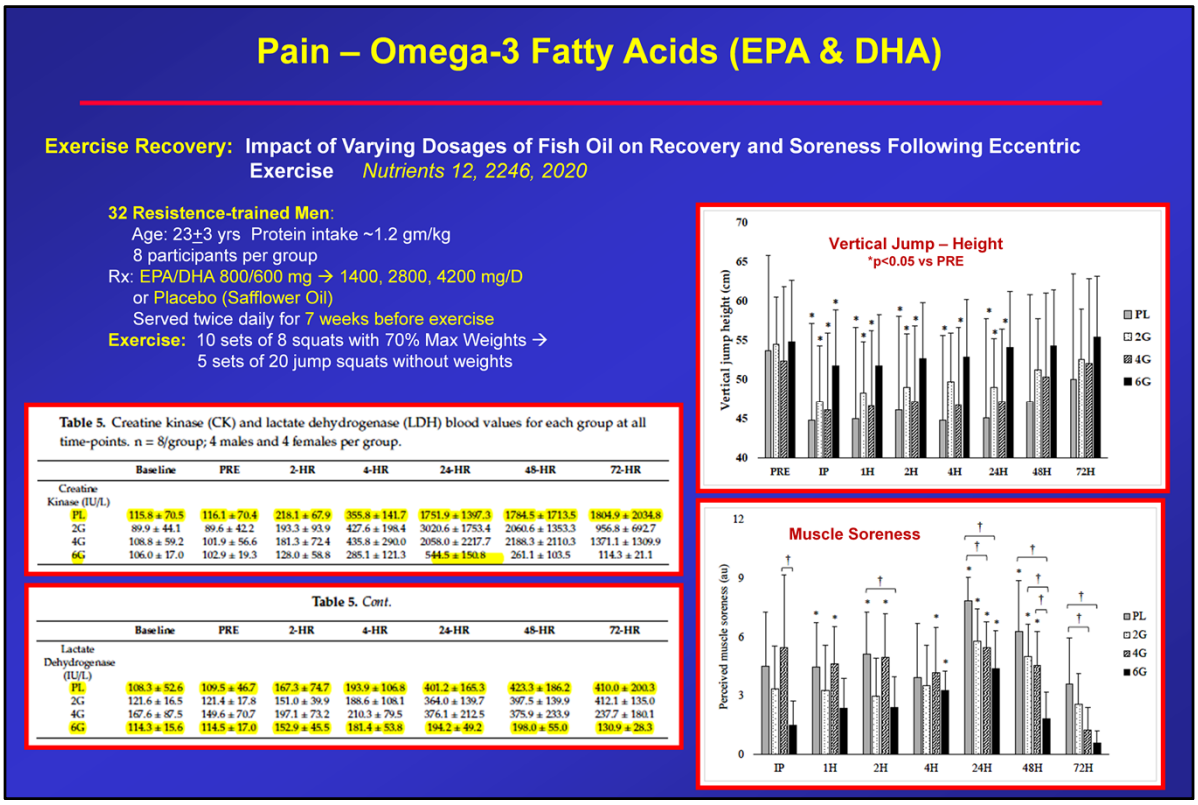
Eight weeks of EPA and DHA supplementation may play a protective role against motor nerve dysfunction and may attenuate muscle damage after eccentric contractions.



Abstract

Evidence suggests that omega-3 fatty acid supplementation could reduce muscle soreness and maintain muscle function following eccentric exercise-induced muscle damage. The aim of this applied field study was to investigate the effectiveness of consuming a protein-based supplement containing 1546 mg of omega-3 polyunsaturated fatty acid (PUFA) (551 mg eicosapentaenoic acid (EPA) and 551 mg docosahexaenoic acid (DHA)) twice daily (FO) compared to a protein-based placebo (P) on muscle soreness, countermovement jump (CMJ) performance and psychological well-being in 20 professional Rugby Union players during 5 weeks of pre-season training. Players completed a 5-point-Likert soreness scale with 5 indicating "no soreness" and a questionnaire assessing fatigue, sleep, stress and mood each morning of training, plus they performed CMJ tests once or twice per week. Data were analysed using magnitude-based inferential statistics and are presented as percent beneficial/trivial/harmful. On day 35, there was a likely (% beneficial/trivial/harmful: 94/5/1) moderate (0.75, standardized mean difference (SMD)) beneficial effect of FO vs. P on the change in lower body muscle soreness compared with day 0 (FO: −3.8 ± 21.7%; P: −19.4 ± 11.2%). There was a likely (92/7/0) moderate (SMD: 0.60) beneficial effect of FO vs. P on CMJ performance (change from baseline to day 35, FO: +4.6 ± 5.9%; P: −3.4 ± 8.6%). From day 20, a moderate beneficial effect of FO on fatigue was observed. In terms of practical relevance, the moderate beneficial effect of adding fish oil to a protein-based

supplement on muscle soreness translated into the better maintenance of explosive power in elite Rugby Union players during pre-season training.



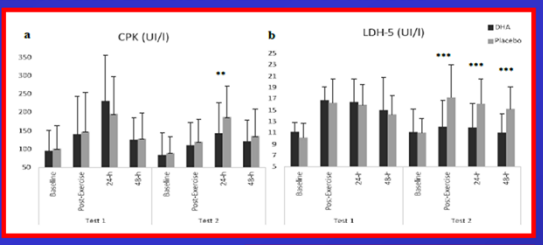
Abstract: Fish oils (FOs) are rich in omega-3 long-chain polyunsaturated fatty acids, which have been purported to enhance recovery of muscular performance and reduce soreness post-exercise. However, the most effective FO dose for optimizing recovery remains unclear. The purpose of this investigation was to examine the effect of FO supplementation dosing on the recovery of measures of muscular performance, perceived soreness, and markers of muscle damage following a rigorous bout of eccentric exercise. Thirty-two college-aged resistance-trained males (~23.6 years, 71.6 kg, 172.1 cm) were supplemented with 2, 4, 6 g/day (G) FO or placebo (PL) for ~7.5 weeks. Following 7 weeks of supplementation, pre-exercise (PRE) performance assessments of vertical jump (VJ), knee extensor strength, 40-yard sprint, T-test agility, and perceived soreness were completed prior to a bout of muscle-damaging exercise and were repeated immediately post (IP), 1-, 2-, 4-, 24-, 48-, and 72-h (H) post-exercise. Repeated measures analysis of variance indicated a treatment time interaction ($p < 0.001$) for VJ and perceived soreness, but no group differences were observed at any time point. VJ returned to PRE (54.8 $\pm$ 7.9 cm) by 1H (51.8 $\pm$ 6.5 cm, $p = 0.112$) for 6G, while no other groups returned to baseline until 48H. Lower soreness scores were observed in 6G compared to PL at 2H (mean difference [MD] = 2.74, $p = 0.046$), at 24H (MD: 3.45, $p < 0.001$), at 48H (MD = 4.45, $p < 0.001$), and at 72H (MD = 3.00, $p = 0.003$). Supplementation with 6G of FO optimized the recovery of jump performance and muscle soreness following a damaging bout of exercise.

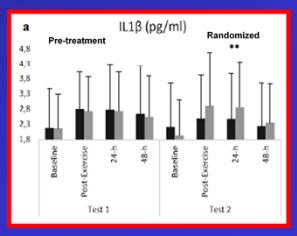
Pain – Omega-3 Fatty Acids (EPA & DHA)

Exercise Recovery: **Supplementation of Re-Esterified Docosahexaenoic and Eicosapentaenoic Acids Reduce Inflammatory and Muscle Damage Markers after Exercise in Endurance Athletes**

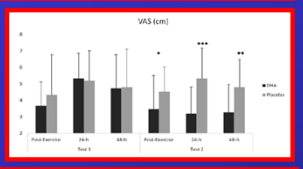
Nutrients 12, 719, 2020

18 healthy, male amateur endurance athletes
Mean Age 36 yo
Exercise: 8 sets of 6 reps ½ squat at 110% of 1-RM
Test: 8 sets of 10 reps of vert jump
Rx: 2.1 gm DHA + 240 mg EPA for 10 weeks vs Plac





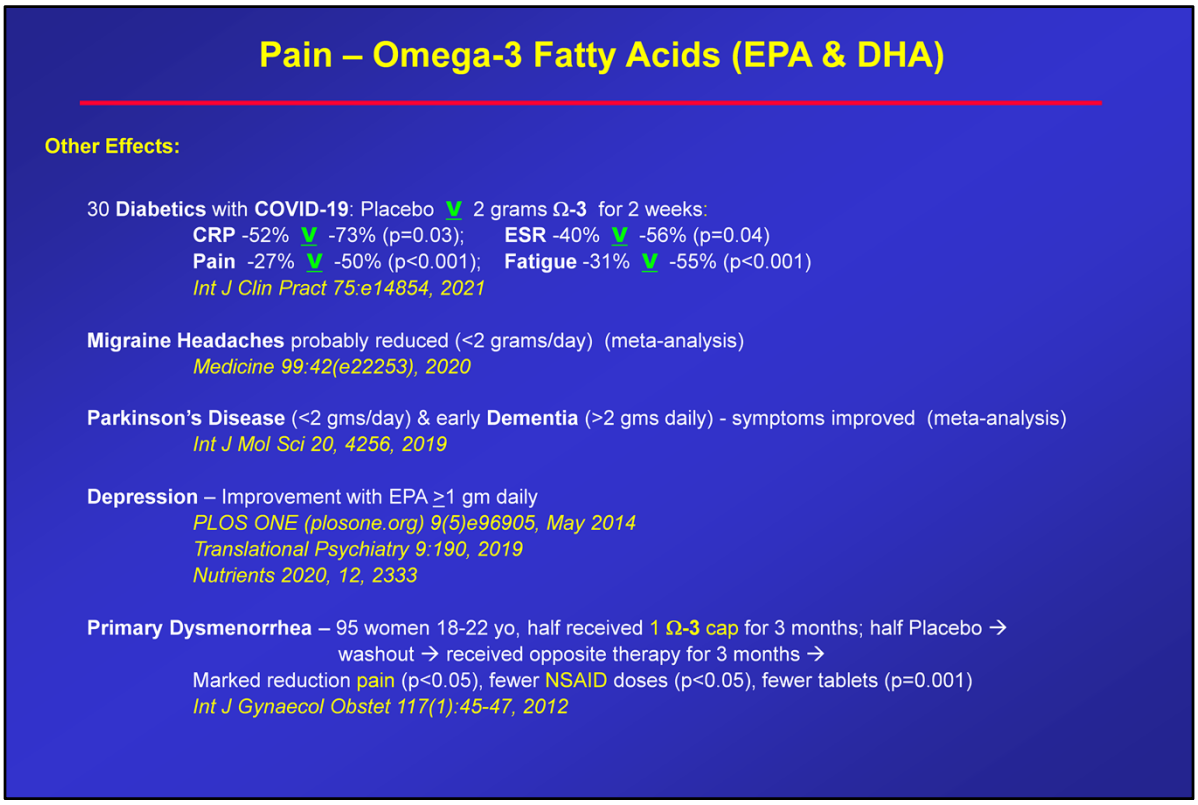
Each subject was tested 4 times:
1. Pre-treatment (Test 1)
2. Randomized & retested after 10 weeks (Test 2)
3. Wash-out for 4 weeks & retested (Test 1)
4. Crossed over & retested (Test 2)



Outcome	Testing Session	Baseline		Post-exercise		post 24 h		post 48 h		ANOVA (supplement × test ×time)		Post-hoc	
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	*F*	*p*	Comparison	*p*
IL1β (pg/ml)	Pre-DHA + EPA	2.18	1.29	2.80	1.21	2.78	1.49	2.64	1.59	4.247	0.011	DHA + EPA test 1 vs. DHA + EPA test 2 (post-exerc)	**0.036**
	Post-DHA + EPA	2.22	1.44	2.49	1.42	2.48	1.48	2.24	1.40			DHA + EPA test 1 vs. DHA + EPA test 2 (24 h)	**0.002**
	Pre-Placebo	2.17	1.12	2.73	1.12	2.73	1.29	2.54	1.36			DHA + EPA test 1 vs. DHA + EPA test 2 (48 h)	**0.004**
	Post-Placebo	1.94	1.17	2.90	1.78	2.86	1.50	2.35	1.28				
IL6 (pg/ml)	Pre-DHA + EPA	2.55	1.29	3.53	1.45	3.12	1.50	2.74	1.55	5.909	0.009	DHA + EPA test 1 vs. DHA + EPA test 2 (post-exerc)	0.062
	Post-DHA + EPA	2.75	1.70	3.07	1.77	2.80	1.76	2.98	2.00			DHA + EPA test 1 vs. DHA + EPA test 2 (24 h)	0.067
	Pre-Placebo	3.02	1.71	3.95	1.89	3.64	1.97	3.07	1.66				
	Post-Placebo	2.66	1.71	3.89	2.32	3.51	2.18	3.16	1.94				

Abstract: This study aimed to analyze the effect of 10 weeks of a highly concentrated docosahexaenoic acid (DHA) + eicosapentaenoic (EPA) supplementation (ratio 8:1) on strength deficit and inflammatory and muscle damage markers in athletes. Fifteen endurance athletes participated in the study. In a randomized, double-blinded cross-over controlled design, the athletes were supplemented with a re-esterified triglyceride containing 2.1 g/day of DHA + 240 mg/day of EPA or placebo for 10 weeks. After a 4-week wash out period, participants were supplemented with the opposite treatment. Before and after each supplementation period, participants performed one eccentric-induced muscle damage exercise training session (ECC). Before, post-exercise min and 24 and 48 h after exercise, muscle soreness, knee isokinetic strength and muscle damage and inflammatory markers were tested. No significant differences in strength deficit variables were found between the two conditions in any of the testing sessions. However, a significant effect was observed in IL-1 ($p = 0.011$) and IL-6 ($p = 0.009$), which showed significantly lower values after DHA consumption than after placebo ingestion. Moreover, a significant main effect was observed in CPK ($p = 0.014$) and LDH-5 ($p = 0.05$), in which significantly lower values were found after DHA + EPA consumption. In addition, there was a significant effect on muscle soreness ($p = 0.049$), lower values being obtained after DHA + EPA consumption. Ten weeks of re-esterified DHA + EPA promoted lower concentrations of inflammation and muscle damage markers and decreased muscle soreness but did not improve the strength

deficit after an ECC in endurance athletes.



Abstract *Int J Clin Pract 75:e14854, 2021*

Aims: We hypothesized that omega-3 fatty acids would be an appropriate adjunct therapy for alleviating the inflammatory response and clinical manifestation in hospitalized patients with Covid-19 disease.

Methods: This was a single-blind randomized controlled trial in Amir-Alam hospital in Tehran. Thirty adult men and women diagnosed with Covid-19 were allocated to either control group (receiving Hydroxychloroquine) or intervention group (receiving Hydroxychloroquine plus 2 grams of Docosahexaenoic acid [DHA] + Eicosapentaenoic acid [EPA]) for 2 weeks. Primary outcome of the intervention including C-reactive protein (CRP), erythrocyte sedimentation rate (ESR) as well as clinical symptoms including body pain, fatigue, appetite and olfactory and secondary outcomes including liver enzymes were determined at the baseline and after omega-3 supplementation. Clinical signs were measured using self-reported questionnaires. There were commercial kits for determination of CRP and liver enzymes concentrations in the serum of patients. For determination of ESR automated hematology analyzer was applied.

Results: In comparison to control group, patients receiving omega-3 indicated favourable changes in all clinical symptoms except for olfactory ($P < .001$ for body pain and fatigue, $P = .03$ for appetite and $P = .21$ for olfactory). Reducing effects of omega-3 supplementation compared with control group were also observed in the levels of ESR and CRP after treatment ($P < .001$ for CRP and $P = .02$ for ESR).

However, no between group differences in the liver enzymes serum concentrations were observed after supplementation ($P > .05$).
Conclusion: Current observations are very promising and indicate that supplementation with moderate dosages of omega-3 fatty acids may be beneficial in the management of inflammation-mediated clinical symptoms in Covid-19 patients.

Abstract *Medicine 99:42(e22253), 2020*
Background: Omega-3 fatty acids (FAs) can produce several beneficial effects and are commonly used for the treatment of migraine symptoms. Although current therapeutic measures for migraine included pharmacological therapies, dietary supplements, and herbal ingredients, dietary patterns, acupuncture, relaxation techniques, biofeedback, and psychotherapy, omega-3 FAs therapeutic role seems to be obtained through the inhibition or reduction of the release of inflammatory cytokines. The present review aims to provide updated information about the effects of omega-3 FAs in migraine treatment, investigating their clinical effects alone or in combination with other substances.
Methods: Bibliographic research was conducted by examining scientific literature from January 2000 until January 31, 2020. Ten clinical studies were included in the review. Quality assessment of randomized controlled trials was performed by using the JADAD scale.
Results: Clinical studies methodology is not always of good quality and results show moderate evidence concerning the therapeutic role of omega-3 FAs in migraine.
Conclusion: Further clinical trials are necessary to implement the knowledge concerning the use of omega-3 fatty acids in the treatment of migraine.

Abstract *Int J Mol Sci 20, 4256, 2019*
A nutritional approach could be a promising strategy to prevent or slow the progression of neurodegenerative diseases such as Parkinson's and Alzheimer's disease, since there is no effective therapy for these diseases so far. The beneficial effects of omega-3 fatty acids are now well established by a plethora of studies through their involvement in multiple biochemical functions, including synthesis of anti-inflammatory mediators, cell membrane fluidity, intracellular signaling, and gene expression. This systematic review will consider epidemiological studies and clinical trials that assessed the impact of supplementation or dietary intake of omega-3 polyunsaturated fatty acids on neurodegenerative diseases such as Parkinson's and Alzheimer's diseases. Indeed, treatment with omega-3 fatty acids, being safe and well tolerated, represents a valuable and biologically plausible tool in the management of neurodegenerative diseases in their early stages.

Abstract *PLOS ONE (plosone.org) 9(5)e96905, May 2014*
Background: Despite omega-3 polyunsaturated fatty acids (PUFA) supplementation

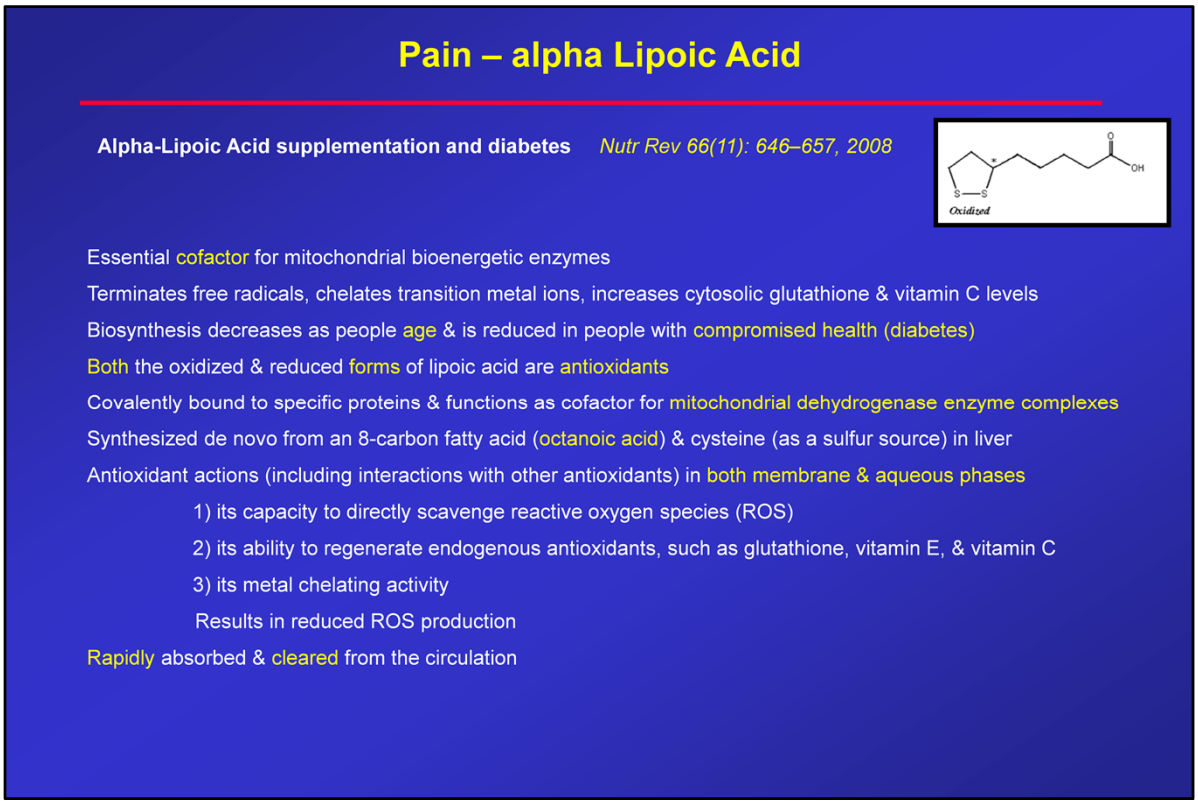
in depressed patients have been suggested to improve depressive symptomatology, previous findings are not univocal.
Objectives: To conduct an updated meta-analysis of randomized controlled trials (RCTs) of omega-3 PUFA treatment of depressive disorders, taking into account the clinical differences among patients included in the studies.
Methods: A search on MEDLINE, EMBASE, PsycInfo, and the Cochrane Database of RCTs using omega-3 PUFA on patients with depressive symptoms published up to August 2013 was performed. Standardized mean difference in clinical measure of depression severity was primary outcome. Type of omega-3 used (particularly eicosapentaenoic acid [EPA] and docosahexaenoic acid [DHA]) and omega-3 as mono- or adjuvant therapy was also examined. Meta-regression analyses assessed the effects of study size, baseline depression severity, trial duration, dose of omega-3, and age of patients.
Results: Meta-analysis of 11 and 8 trials conducted respectively on patients with a DSM-defined diagnosis of major depressive disorder (MDD) and patients with depressive symptomatology but no diagnosis of MDD demonstrated significant clinical benefit of omega-3 PUFA treatment compared to placebo (standardized difference in random-effects model 0.56 SD [95% CI: 0.20, 0.92] and 0.22 SD [95% CI: 0.01, 0.43], respectively; pooled analysis was 0.38 SD [95% CI: 0.18, 0.59]). Use of mainly EPA within the preparation, rather than DHA, influenced final clinical efficacy. Significant clinical efficacy had the use of omega-3 PUFA as adjuvant rather than mono-therapy. No relation between efficacy and study size, baseline depression severity, trial duration, age of patients, and study quality was found. Omega-3 PUFA resulted effective in RCTs on patients with bipolar disorder, whereas no evidence was found for those exploring their efficacy on depressive symptoms in young populations, perinatal depression, primary disease other than depression and healthy subjects.
Conclusions: The use of omega-3 PUFA is effective in patients with diagnosis of MDD and on depressive patients without diagnosis of MDD.

Abstract *Translational Psychiatry 9:190, 2019*
We conducted this meta-analysis of double-blind randomized placebo-controlled trials to estimate the efficacy of omega-3 polyunsaturated fatty acids (PUFAs), especially docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA), in the improvement of depression. We applied a systematic bibliographic search in PubMed and EMBASE for articles published prior to 20 December 2017. This meta-analysis was performed using RevMan 5.3 and R 3.4.3, and means and standard deviations were calculated in fixed- or random-effects models based on the results of the Q-test. A sensitivity analysis was also conducted to evaluate the stability of the results, and publication bias was evaluated by a funnel plot and Egger's linear regression analysis. Our search resulted in 180 articles; we analyzed 26 studies, which included 2160 participants. The meta-analysis showed an overall beneficial effect of omega-3 polyunsaturated

fatty acids on depression symptoms (SMD = −0.28, P = 0.004). Compared with placebo, EPA-pure (=100% EPA) and EPA-major formulations (≥60% EPA) demonstrated clinical benefits with an EPA dosage ≤1 g/d (SMD = −0.50, P = 0.003, and SMD = −1.03, P = 0.03, respectively), whereas DHA-pure and DHA-major formulations did not exhibit such benefits. Current evidence supports the finding that omega-3 PUFAs with EPA ≥ 60% at a dosage of ≤1 g/d would have beneficial effects on depression. Further studies are warranted to examine supplementation with omega-3 PUFAs for specific subgroups of subjects with inflammation, severity of depression, and the dose response for both EPA and DHA supplementation.

Abstract *Nutrients 12:2333, 2020*
Most of the global population is deficient in long-chain marine omega-3s. In particular, docosahexaenoic acid (DHA), a long-chain omega-3 fatty acid, is important for brain and eye development. Additionally, DHA plays a significant role in mental health throughout early childhood and even into adulthood. In the brain, DHA is important for cellular membrane fluidity, function and neurotransmitter release. Evidence indicates that a low intake of marine omega-3s increases the risk for numerous mental health issues, including Attention Deficit Hyperactivity Disorder (ADHD), autism, bipolar disorder, depression and suicidal ideation. Studies giving supplemental marine omega-3s have shown promise for improving numerous mental health conditions. This paper will review the evidence surrounding marine omega-3s and mental health conditions.

Abstract *Int J Gynaecol Obstet 117(1):45-47, 2012*
Objective: To examine whether dietary supplementation with omega-3 fatty acids relieved symptoms of primary dysmenorrhea.
Methods: Women aged 18-22 years with primary dysmenorrhea were enrolled in a double-blind crossover study. Women assigned to group 1 (n=47) received 1 omega-3 capsule daily for 3 months, followed by placebo for 3 months. Women in group 2 (n=48) received placebo for 3 months, followed by omega-3 for 3 months. A washout period was performed in both groups. Participants used 400 mg of ibuprofen as a rescue dose if severe menstrual pains were experienced.
Results: A marked reduction in pain intensity was observed after 3 months of treatment with omega-3 fatty acids (P<0.05). Women who received omega-3 fatty acids required fewer rescue doses than women who received placebo (P<0.05). The mean numbers of ibuprofen tablets used after 3 months with omega-3 fatty acids were 4.3±2.1 (group 1) and 3.2±2.5 (group 2); the mean numbers of tablets used after 3 months of placebo were 5.3±2.2 (group 1) and 6.0±2.6 (group 2) (P=0.001 for both).
Conclusion: Supplementation with omega-3 fatty acids reduced the symptom intensity of primary dysmenorrhea. Supplementation efficacy was sufficient to decrease the ibuprofen rescue dose.



Abstract

Diabetes is a common metabolic disorder that is usually accompanied by increased production of reactive oxygen species or by impaired antioxidant defenses. Importantly, oxidative stress is particularly relevant to the risk of cardiovascular disease. Alpha-lipoic acid (LA), a naturally occurring dithiol compound, has long been known as an essential cofactor for mitochondrial bioenergetic enzymes. LA is a very important micronutrient with diverse pharmacologic and antioxidant properties. Pharmacologically, LA improves glycemic control and polyneuropathies associated with diabetes mellitus; it also effectively mitigates toxicities associated with heavy metal poisoning. As an antioxidant, LA directly terminates free radicals, chelates transition metal ions, increases cytosolic glutathione and vitamin C levels, and prevents toxicities associated with their loss. These diverse actions suggest that LA acts by multiple mechanisms both physiologically and pharmacologically. Its biosynthesis decreases as people age and is reduced in people with compromised health, thus suggesting a possible therapeutic role for LA in such cases. Reviewed here is the known efficacy of LA with particular reference to types 1 and 2 diabetes. Particular attention is paid to the potential benefits of LA with respect to glycemic control, improved insulin sensitivity, oxidative stress, and neuropathy in diabetic patients. It appears that the major benefit of LA supplementation is in patients with diabetic neuropathy.

INTRODUCTION

Alpha-lipoic acid (LA) also known as thioctic acid, was first isolated from bovine liver in 1950. Lipoic acid contains two thiol groups, which may be oxidized or reduced. As with the thiol antioxidant glutathione, LA is part of a redox pair, being the oxidized partner of the reduced form dihydrolipoic acid (DHLA). Unlike glutathione, for which only the reduced form is an antioxidant, both the oxidized and reduced forms of lipoic acid are antioxidants. LA is 6,8-dithio-octanoic acid, an eight-carbon disulphide containing a single chiral center (Figure 1). LA also contains an asymmetric carbon, thus resulting in two possible optical isomers (RLA and S-LA). Only the R-isomer is endogenously synthesized and bound to protein. Lipoic acid supplements may contain either R-LA or a 50/50 (racemic) mixture of R-LA and S-LA. LA is reduced in vivo to its dithiol form, DHLA, which also possesses biological activity. Lipoic acid is a naturally occurring compound that is synthesized in small amounts by plants and animals, including humans. Endogenously synthesized LA is covalently bound to specific proteins, which function as cofactors for mitochondrial dehydrogenase enzyme complexes. In addition to the physiological functions of protein-bound LA, there is an increasing scientific and medical interest in potential therapeutic uses of pharmacological doses of free LA. Considering its role in biochemical processes, lipoic acid was initially included in the vitamin B complex. However, at present, LA is not considered to be a vitamin.

METABOLISM AND BIOAVAILABILITY

LA is synthesized de novo from an 8-carbon fatty acid (octanoic acid) and cysteine (as a sulfur source) in liver. Its catabolism also takes place in liver. Due to an asymmetric carbon having four different attached groups, LA exists as two enantiomers: the R-enantiomer and the Senantiomer. Naturally occurring lipoic acid is the R-form, but synthetic lipoic acid is a racemic mixture of R-form and S-form. Both forms seem to have different potencies; it was previously shown that the R-form is more potent than the S-form in its ability to stimulate glucose uptake in L6 myotubes,5 as well as to increase insulin-stimulated glucose uptake in obese Zucker rats. On the other hand, the S-form exerts a slightly increased affinity for glutathione reductase. Thus, the two forms of LA differ in the potency in which they exert the various biological activities of this compound.

LIPOIC ACID AS A PRIMARY ANTIOXIDANT

As stated by Packer et al., “an ideal therapeutic antioxidant should fulfill several criteria. These include absorption from the diet, conversion in cells and tissues into usable form, a variety of antioxidant actions (including interactions with other antioxidants) in both membrane and aqueous phases, and low toxicity.” LA is unique among natural antioxidants in its ability to fulfill all of these requirements, making it a potentially highly effective therapeutic agent for a number of conditions in which

oxidative damage has been implicated. LA's antioxidant properties consist of the following: 1) its capacity to directly scavenge reactive oxygen species (ROS); 2) its ability to regenerate endogenous antioxidants, such as glutathione and, vitamins E and C; and 3) its metal chelating activity, resulting in reduced ROS production.

Scavenging reactive oxygen and nitrogen species

ROS and reactive nitrogen species (RNS) are highly reactive compounds with the potential to damage DNA, proteins, and lipids in cell membranes. Common antioxidants are either water soluble or lipid membrane-soluble agents. In contrast, LA has both hydrophilic and hydrophobic properties. This amphiphilic characteristic of LA is unique among antioxidants. LA can therefore elicit its antioxidant action in the cytosol as well as in the plasma membrane (aqueous and lipid media of the cell), and in serum and lipoproteins (aqueous and lipid media of blood) in contrast to vitamin C (which is hydrophilic) and vitamin E (which is hydrophobic). The highest tissue concentrations of free LA likely achieved by oral supplementation are at least 10 times lower than those of other intracellular antioxidants, such as vitamin C and glutathione because of its rapid clearance rate. Earlier, our group9 reported that LA supplementation (600 mg/day for 2 months) in healthy volunteers significantly increased the lag time of LDL lipid peroxide formation for both copper-catalyzed and 2,2′-azobis(2-amidinopropanane) dihydrochloride (AAPH)-induced LDL oxidation, decreased urinary F2-isoprostane levels, and plasma carbonyl levels after AAPH oxidation.

Regeneration of other antioxidants

When an antioxidant scavenges a free radical, it becomes oxidized itself and is not able to scavenge additional ROS or RNS until it has been reduced. DHLA is a potent reducing agent with the capacity to reduce the oxidized forms of several important antioxidants, including vitamin C and glutathione. In general, DHLA has superior antioxidant activity to LA. DHLA can regenerate vitamin C and vitamin E from their oxidized forms. Although reduced glutathione has twice the chemical reactivity in its thiol group, DHLA is superior to glutathione in regenerating vitamin C. DHLA may also reduce the oxidized form of alpha-tocopherol (the alpha-tocopheryl radical) directly or indirectly, by reducing the oxidized form of vitamin C (dehydroascorbate), which is able to reduce the alpha-tocopheryl radical. Coenzyme Q10 is an important component of the mitochondrial electron transport chain that has antioxidant activity as well. DHLA can also reduce oxidized forms of coenzyme Q10, which may additionally reduce the alpha-tocopheryl radical. Thus, LA also plays an important role in the synergism of antioxidants described as the body's "antioxidant network" by Packer et al. It directly recycles and extends the metabolic lifespans of vitamin C, glutathione, and coenzyme Q10, and it indirectly renews vitamin E. However, in the study reported by our group, LA supplementation did not produce any change in the

levels of plasma or LDL alpha-tocopherol, though the baseline levels of AT were not deficient in the study population. Therefore, it was concluded that LA does not appear to act through regeneration of AT levels. Furthermore, LA has been reported to increase glutathione synthesis in aged animals by increasing the expression of gamma-glutamylcysteine ligase, the rate-limiting enzyme in glutathione synthesis, and by increasing cellular uptake of cysteine, an amino acid required for glutathione synthesis. Overall, based on all the above-stated evidence, it is unclear if LA really plays a part in the control of cellular redox status.

Metal chelation

Redox-active metal ions, such as free iron and copper, can induce oxidative damage by catalyzing reactions that generate highly reactive free radicals. Compounds that chelate
(bind) free metal ions in a way that prevents them from generating free radicals offer promise in the treatment of chronic diseases in which metal-induced oxidative damage may play a role. Both LA and DHLA have been found to inhibit copper- and iron-mediated oxidative damage in the test tube and to inhibit excess iron and copper accumulation in animal models.

LIPOIC ACID AVAILABILITY AND SUPPLEMENTS

In humans, LA is synthesized in liver and other tissues and is also obtained from both animal and plant sources in the diet. LA is readily absorbed from the diet and is rapidly converted to DHLA by reduced nicotinamide adenine dinucleotide or reduced nicotinamide adenine dinucleotide phosphate in most tissues.

Food sources

R-LA occurs naturally in foods covalently bound to lysine in proteins (lipoyllysine). High concentrations of LA are found in animal tissues with extensive metabolic activity such as
heart, liver, and kidney. The plant sources of LA, listed from highest to lowest, are spinach, broccoli, tomatoes, garden peas, Brussel sprouts, and rice bran.

Supplements

Unlike LA in foods, LA in supplements is free; thus, it is not bound to protein. Moreover, the amounts of LA available in dietary supplements (200–600 mg) are likely to be as much as 1000 times greater than the amounts that could be obtained from the diet. In Germany, LA is approved for the treatment of diabetic neuropathies and is available by prescription. In the United States, LA is available over the counter as a dietary supplement. Food intake is reported to reduce the bioavailability of LA. Therefore, LA is generally recommended to be taken up on an empty stomach (1 h before or 2 h after eating).

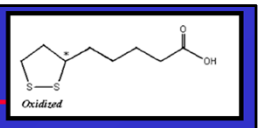
Racemic versus R-LA supplements

R-LA is the isomer that is synthesized by plants and animals and functions as a cofactor for mitochondrial enzymes in its protein-bound form. Direct comparisons of the bioavailability of oral racemic LA and R-LA supplements have not been published. After oral dosing with racemic LA, peak plasma concentrations of R-LA were found to be 40–50% higher than SLA, suggesting R-LA is better absorbed than S-LA, but both isomers are rapidly metabolized and eliminated. However, virtually all of the published studies of LA supplementation in humans have used racemic LA. At present, it is not clear whether R-LA supplements are more effective than racemic LA supplements in humans.

LIPOIC ACID CIRCULATING VALUES AND DEFICIENCY IN TISSUES

Endogenous levels of plasma LA are reported to be 1–25 ng/mL and of DHLA as 33–145 ng/mL in healthy human volunteers. Overall, humans are able to synthesize enough LA to meet their needs for enzyme cofactors. However, its synthesis declines with age and in people with compromised health including diabetes and associated abnormalities, such as diabetic neuropathy. Thus, in these cases, LA may need to be obtained from outside sources by consuming certain foods and from supplements.

Pain – alpha Lipoic Acid



Nerve Conduction: Treatment of diabetic polyneuropathy with the antioxidant thioctic acid (α-lipoic acid): A two year multicenter randomized double-blind placebo-controlled trial (ALADIN II)
Free Radical Research, 31:3, 171-179, 1999

TABLE II Changes of peripheral nerve function indices from baseline to 24 month

65 patients	Placebo	TA 600 mg/day	TA 1200
Sural SNCV (m/s)			
Baseline	41.6 ± 4.1	41.0 ± 7.1	38.9 ± 95
Change (month 24)	−0.1 ± 4.8	3.0 ± 3.0[a]	3.8 ± 4.2[a]
Sural SNAP (μV)			
Baseline	33 ± 3.2	3.1 ± 2.4	25 ± 2.0
Change (month 24)	−0.7 ± 1.5	0.3 ± 1.4[a]	0.6 ± 2.5[b]
Tibial MNCV (m/s)			
Baseline	48.2 ± 6.2	46.6 ± 6.2	47.1 ± 5.1
Change (month 24)	−1.5 ± 2.9	−0.3 ± 5.2	1.2 ± 3.8[a]
Tibial nerve DML (ms)			
Baseline	5.4 ± 1.3	5.1 ± 1.2	5.2 ± 0.9
Change (month 24)	0.0 ± 1.1	−0.03 ± 1.1	−0.17 ± 1.3

Values are mean ± SD, [a]$p < 0.05$ vs PLA, [b]$p = 0.076$ vs PLA.
SNCV: sensory nerve conduction velocity; MNCV: motor nerve conduction velocity; SNAP: sensory nerve action potential; DML: distal motor latency.

Free Radical Research, 31:3, 171-179, 1999

ABSTRACT: Short-term trials with the antioxidant thioctic acid (TA) appear to improve neuropathic symptoms in diabetic patients, but the long-term response remains to be established. Therefore, Type 1 and Type 2 diabetic patients with symptomatic polyneuropathy were randomly assigned to three treatment regimens: (1) 2 x 600mg of TA (TA 1200), (2) 600mg of TA plus placebo (PLA) (TA 600) or (3) placebo and placebo (PLA). A trometamol salt solution of TA of 1200 or 600 mg or PLA was intravenously administered once daily for five consecutive days before enrolling the patients in the oral treatment phase. The study was prospective, PLA-controlled, randomized, double-blind and conducted for two years. Severity of diabetic neuropathy was assessed by the Neuropathy Disability Score (NDS) and electrophysiological attributes of the sural (sensory nerve conduction velocity (SNCV), sensory nerve action potential (SNAP)) and the tibia (motor nerve conduction velocity (MNCV), motor nerve distal latency (MNDL)) nerve. Statistical analysis was performed after independent reviewers excluded all patients with highly variable data allowing a final analysis of 65 patients (TA 1200: n = 18, TA 600: n =27; PLA: n = 20).
At baseline no significant differences were noted between the groups regarding the demographic variables and peripheral nerve function parameters for these 65 patients. Statistically significant changes after 24 months between TA and PLA were observed (mean ± SD) for

sural SNCV: + 3.8 ± 4.2 m/s in TA 1200, +3.0±3.0 m/s in TA 600, - 0.1+ 4.8 m/s in PLA ($p < 0.05$ for TA 1200 and TA 600 vs. PLA);
sural SNAP: + 0.6 ± 2.5 microV in TA 1200, + 0.3 ± 1.4 microV in TA 600, - 0.7 ± 1.5 microV in PLA ($p = 0.076$ for TA 1200 vs. PLA and $p < 0.05$ for TA 600 vs. PLA), and in
tibial MNCV: + 1.2 ± 3.8 m/s in TA 1200, **-** 0.3 ± 5.2 m/s in TA 600, - 1.5 ± 2.9 m/s in PLA ($p < 0.05$ for TA 1200 vs. PLA).
No significant differences between the groups after 24 months were noted regarding the tibial MNDL and the NDS.
We conclude that in a subgroup of patients after exclusion of patients with excessive test variability throughout the trial, TA appeared to have a beneficial effect on several attributes of nerve conduction.

Pain – alpha Lipoic Acid

Neuropathy Symptoms: Oral Treatment With α-Lipoic Acid Improves Symptomatic Diabetic Polyneuropathy
The SYDNEY 2 trial *Diabetes Care 29:2365–2370, 2006*

Table 2—*Baseline levels and changes from baseline (negative values correspond to improvement) in the TSS and its individual subscores after 5 weeks of treatment (last value carried forward)*

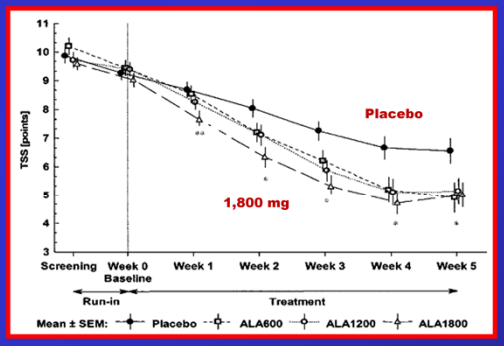
181 patients		Placebo	ALA600 mg/day	ALA1200	ALA1800
TSS	Total Symptom Score				
Baseline		9.27 ± 1.56	9.44 ± 1.86	9.40 ± 1.64	9.02 ± 1.61
Change		−2.92 ± 3.18	−4.85 ± 3.03*	−4.50 ± 3.28*	−4.70 ± 3.54*
Stabbing pain					
Baseline		2.21 ± 0.77	2.32 ± 0.94	2.38 ± 0.89	2.03 ± 0.88
Change		−0.83 ± 1.14	−1.40 ± 1.15*	−1.56 ± 1.07*	−1.46 ± 1.20*
Burning pain					
Baseline		2.11 ± 0.87	2.21 ± 1.07	2.17 ± 1.05	2.15 ± 1.03
Change		−0.50 ± 1.15	−1.32 ± 1.07*	−1.09 ± 1.19*	−1.15 ± 1.41*
Paresthesia					
Baseline		2.21 ± 0.63	2.32 ± 0.80	2.12 ± 0.80	2.17 ± 0.69
Change		−0.80 ± 1.17	−1.16 ± 1.26	−0.85 ± 1.21	−1.12 ± 1.20
Numbness					
Baseline		2.74 ± 0.67	2.58 ± 0.67	2.73 ± 0.66	2.67 ± 0.72
Change		−0.79 ± 1.09	−0.97 ± 1.06	−0.99 ± 1.13	−0.98 ± 1.16

Data are means ± SD. *P < 0.05 vs. placebo.

Table 3—*Screening levels and changes from screening in NSC scores, NIS, and $NIS_{[LL]}$ after 5 weeks of treatment (last value carried forward)*

		Placebo	ALA600	ALA1200	ALA1800
NSC number	Neuropathy Symptoms & Change				
Screening		6.8 ± 1.7	7.0 ± 1.6	7.1 ± 1.5	6.6 ± 1.5
Change		−1.7 ± 2.1	−2.8 ± 2.1*	−2.8 ± 2.2*	−2.7 ± 2.5†
NSC severity					
Screening		14.1 ± 4.3	14.4 ± 4.4	14.7 ± 4.5	13.5 ± 3.5
Change		−4.9 ± 4.3	−7.4 ± 4.6*	−7.2 ± 5.0*	−7.6 ± 4.2*
NSC change					
Screening		−0.1 ± 0.7	0.0 ± 0.3	−0.2 ± 0.6	0.0 ± 0.3
Change		+5.4 ± 4.6	+8.6 ± 5.6*	+8.5 ± 5.3*	+9.5 ± 5.1*
NIS	Neuropathy Impairment Score [Lower Limbs]				
Screening		19.2 ± 17.1	19.4 ± 17.6	18.7 ± 14.1	17.6 ± 15.8
Change		−2.38 ± 6.06	−3.80 ± 4.61	−3.85 ± 4.57*	−3.85 ± 6.49†
$NIS_{[LL]}$					
Screening		14.5 ± 14.6	15.4 ± 14.4	15.0 ± 11.1	13.0 ± 11.6
Change		−2.08 ± 5.57	−3.75 ± 4.41§	−2.63 ± 3.28	−2.70 ± 5.33
$NIS_{[LL]}$ sensory function					
Screening		7.05 ± 3.21	6.80 ± 3.76	7.21 ± 2.82	6.53 ± 2.79
Change		−1.05 ± 1.99	−2.25 ± 2.27*	−1.73 ± 1.72‖	−1.55 ± 1.81

Data are means ± SD. Negative values correspond to improvement. *P < 0.05, †P = 0.08, ‡P = 0.055, §P = 0.07, and ‖P = 0.09 (each vs. placebo).



Abstract *Diabetes Care 29:2365–2370, 2006*

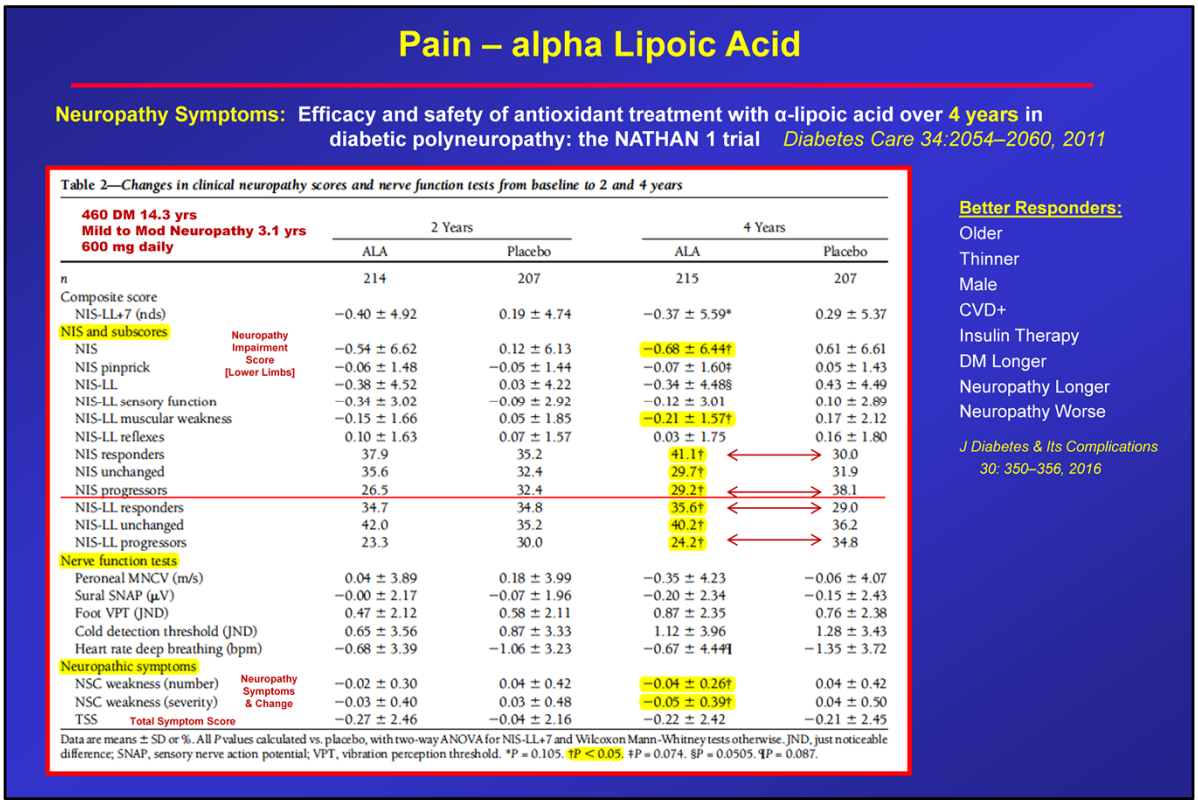
Objective: The aim of this trial was to evaluate the effects of alpha-lipoic acid (ALA) on positive sensory symptoms and neuropathic deficits in diabetic patients with distal symmetric polyneuropathy (DSP).

Research design and methods: In this multicenter, randomized, double-blind, placebo-controlled trial, 181 diabetic patients in Russia and Israel received once-daily oral doses of 600 mg (n = 45) (ALA600), 1,200 mg (n = 47) (ALA1200), and 1,800 mg (ALA1800) of ALA (n = 46) or placebo (n = 43) for 5 weeks after a 1-week placebo run-in period. The primary outcome measure was the change from baseline of the Total Symptom Score (TSS), including stabbing pain, burning pain, paresthesia, and asleep numbness of the feet. Secondary end points included individual symptoms of TSS, Neuropathy Symptoms and Change (NSC) score, Neuropathy Impairment Score (NIS), and patients' global assessment of efficacy.

Results: Mean TSS did not differ significantly at baseline among the treatment groups and on average decreased by 4.9 points (51%) in ALA600, 4.5 (48%) in ALA1200, and 4.7 (52%) in ALA1800 compared with 2.9 points (32%) in the placebo group (all P < 0.05 vs. placebo). The corresponding response rates (>/=50% reduction in TSS) were 62, 50, 56, and 26%, respectively. Significant improvements favoring all three ALA groups were also noted for stabbing and burning pain, the NSC score, and the patients' global assessment of efficacy. The NIS was numerically reduced. Safety analysis showed a dose-dependent increase in nausea, vomiting, and

vertigo.

Conclusions: Oral treatment with ALA for 5 weeks improved neuropathic symptoms and deficits in patients with DSP. An oral dose of 600 mg once daily appears to provide the optimum risk-to-benefit ratio.



Abstract *Diabetes Care 34:2054–2060, 2011*

Objective: To evaluate the efficacy and safety of α-lipoic acid (ALA) over 4 years in mild-to-moderate diabetic distal symmetric sensorimotor polyneuropathy (DSPN).

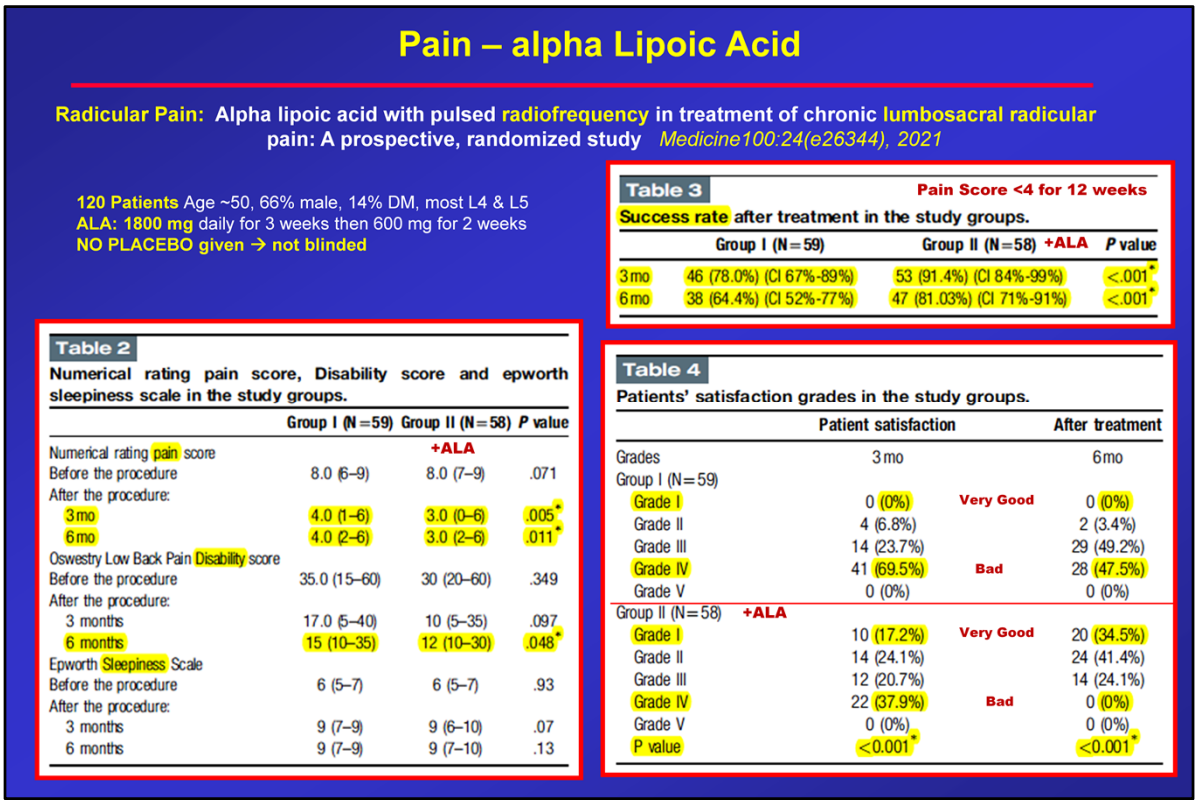
RESEARCH DESIGN AND METHODS In a multicenter randomized double-blind parallel-group trial, 460 diabetic patients with mild-to-moderate DSPN were randomly assigned to oral treatment with 600 mg ALA once daily (n = 233) or placebo (n = 227) for 4 years. Primary end point was a composite score (Neuropathy Impairment Score [NIS]-Lower Limbs [NIS-LL] and seven neurophysiologic tests). Secondary outcome measures included NIS, NIS-LL, nerve conduction, and quantitative sensory tests (QSTs).

Results: Change in primary end point from baseline to 4 years showed no significant difference between treatment groups (P = 0.105). Change from baseline was significantly better with ALA than placebo for NIS (P = 0.028), NIS-LL (P = 0.05), and NIS-LL muscular weakness subscore (P = 0.045). More patients showed a clinically meaningful improvement and fewer showed progression of NIS (P = 0.013) and NIS-LL (P = 0.025) with ALA than with placebo. Nerve conduction and QST results did not significantly worsen with placebo. Global assessment of treatment tolerability and discontinuations due to lack of tolerability did not differ between the groups. The rates of serious adverse events were higher on ALA (38.1%) than on placebo (28.0%).

Conclusions: Four-year treatment with ALA in mild-to-moderate DSPN did not

influence the primary composite end point but resulted in a clinically meaningful improvement and prevention of progression of neuropathic impairments and was well tolerated. Because the primary composite end point did not deteriorate significantly in placebo-treated subjects, secondary prevention of its progression by ALA according to the trial design was not feasible.

Abstract *Journal of Diabetes and Its Complications 30: 350–356, 2016*
Aims: We aimed to analyze the impact of baseline factors on the efficacy of α-lipoic acid (ALA) over 4 years in the NATHAN 1 trial.
Methods: This was a post-hoc analysis of the NATHAN 1 trial, a 4-year randomized study including 460 diabetic patients with mild-to-moderate polyneuropathy using ALA 600 mg qd or placebo. Amongst others, efficacy measures were the Neuropathy Impairment Score of the lower limbs (NIS-LL) and heart rate during deep breathing (HRDB).
Results: Improvement and prevention of progression of NIS-LL (ΔNIS-LL≥2 points) with ALA vs. placebo after 4 years was predicted by higher age, lower BMI, male sex, normal blood pressure, history of cardiovascular disease (CVD), insulin treatment, longer duration of diabetes and neuropathy, and higher neuropathy stage. Participants treated with ALA who received ACE inhibitors showed a better outcome in HRDB after 4 years.
Conclusions: Better outcome in neuropathic impairments following 4-year treatment with α-lipoic acid was predicted by normal BMI and blood pressure and higher burden due to CVD, diabetes, and neuropathy, while improvement in cardiac autonomic function was predicted by ACE inhibitor treatment. Thus, optimal control of CVD risk factors could contribute to improved efficacy of α-lipoic acid in patients with higher disease burden.



Abstract *Medicine100:24(e26344), 2021*

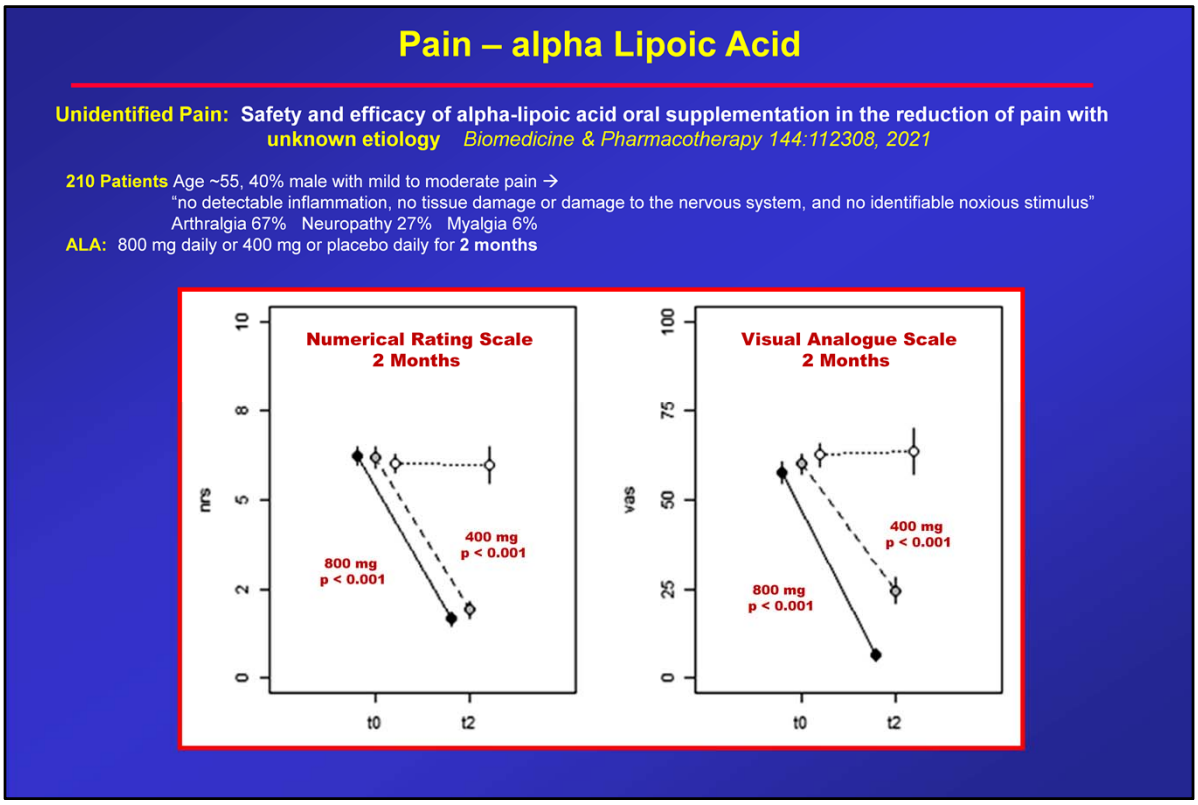
Background: The effect of adding alpha lipoic acid (ALA) to pulsed radiofrequency (PRF) for treatment of lumbar-sacral pain was evaluated.

Objective: To evaluate the effect of using ALA as an adjuvant therapy with PRF for treatment of chronic lumbosacral radicular pain caused by herniated disc.

Methods: One hundred twenty patients with lumbo-sacral radicular pain allocated into 2 groups. Group I: treated with PRF at 42°C for 120 seconds. Group II: treated as in group I, plus oral ALA 600 mg (Thiotacid 600 mg, EVA PHARMA, Egypt) three times per day (1800 mg/day) for 3 weeks then 600 mg once daily for 2 weeks. The lumbo-sacral radicular pain evaluated using the numerical rating pain score and Oswestry Disability Index.

Results: Success rate was significantly higher in group II at 3 and 6 months after intervention. The median values of the numerical rating pain score and the Oswestry Disability Index were significantly lower in group II with no significant difference in Epworth Sleepiness Scale. No major complications were reported in both groups.

Conclusion: The current study supports the use of ALA with PRF on the dorsal root ganglion for treating lumbosacral radicular pain.



ABSTRACT *Biomedicine & Pharmacotherapy 144:112308, 2021*

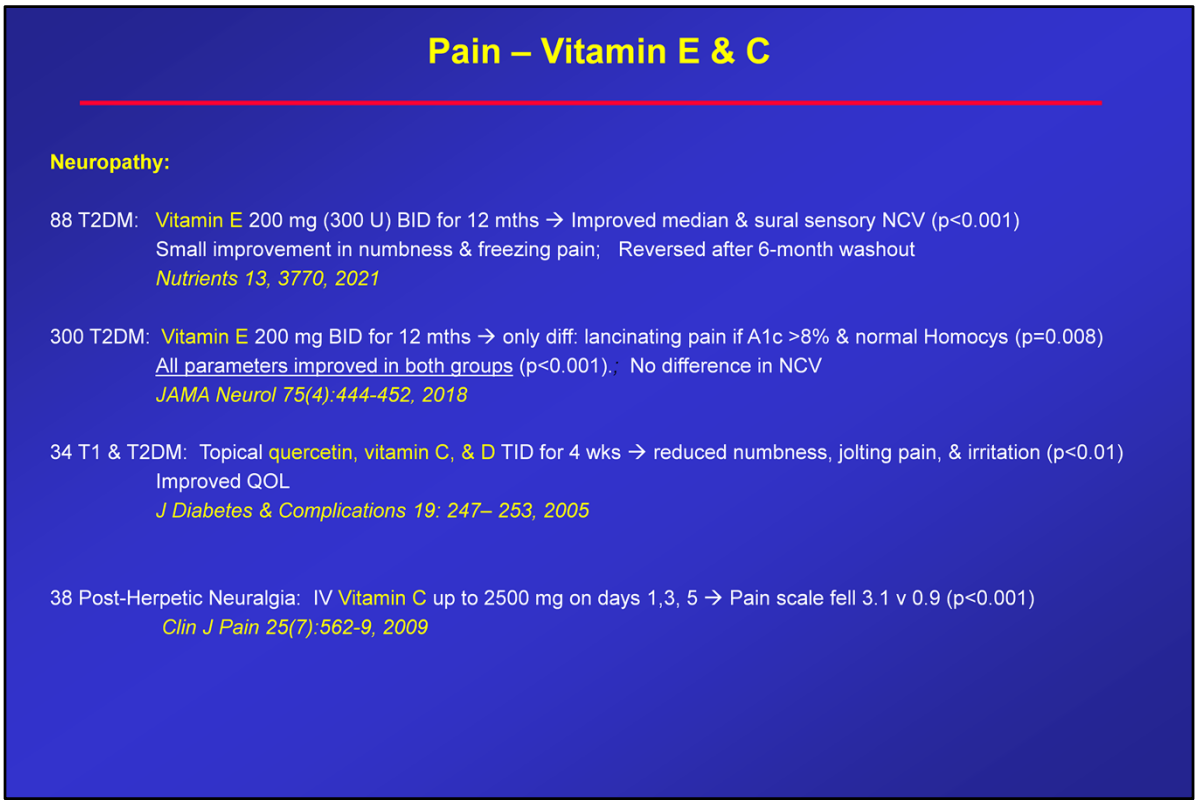
Introduction: Extensive evidence suggests that alpha-lipoic acid (ALA) is effective in diabetic neuropathy pain management. However, little is known on its safety and efficacy in reducing idiopathic pain in normoglycemic subjects. The aim of this study was to evaluate ALA food supplement safety and efficacy in the reduction of different forms of idiopathic pain.

Methods: Two-hundred and ten normoglycemic adults suffering from idiopathic pain (i.e. 57 subjects with primitive neuropathic pain, 141 subjects with arthralgia with unknown etiology, and 12 subjects with idiopathic myalgia) were randomized to receive placebo, 400 mg/day, or 800 mg/day of ALA. Participants underwent two visits (at baseline = t0, and after 2 months = t1) in which two validated questionnaires for pain (numerical rating scale [NRS] and visual analogue scale [VAS]) were collected; fasting blood glucose assessment, adverse effects, and renal and hepatic toxicity were also monitored.

Results: At t1, none of subjects treated with ALA reported a decreased glycemia or adverse effects. The treated subjects showed a significant reduction in NRS ($p < 0.001$) while the placebo group did not show any NRS reduction ($p = 0.86$). Similar results were also obtained for VAS. Statistical analysis aimed at detecting possible differences in NRS and VAS scores among treatment groups based on the source of pain did not reveal any significant effect.

Conclusions: Since the management of idiopathic pain is challenging for

physicians, the use of ALA food supplements could be a feasible option, based on its safety and efficacy compared to commonly-used analgesic drugs.



Nutrients 13, 3770, 2021 **Abstract:** Diabetic peripheral neuropathy (DPN) is the most common microvascular complication of diabetes that affects approximately half of the diabetic population. Up to 53% of DPN patients experience neuropathic pain, which leads to a reduction in the quality of life and work productivity. Tocotrienols have been shown to possess antioxidant, anti-inflammatory, and neuroprotective properties in preclinical and clinical studies. This study aimed to investigate the effects of tocotrienol-rich vitamin E (Tocovid SuprabioTM) on nerve conduction parameters and serum biomarkers among patients with type 2 diabetes mellitus (T2DM). A total of 88 patients were randomized to receive 200 mg of Tocovid twice daily, or a matching placebo for 12 months. Fasting blood samples were collected for measurements of HbA1c, renal profile, lipid profile, and biomarkers. A nerve conduction study (NCS) was performed on all patients at baseline and subsequently at 2, 6, 12 months. Patients were reassessed after 6 months of washout.
After 12 months of supplementation, patients in the Tocovid group exhibited highly significant improvements in conduction velocity (CV) of both median and sural sensory nerves as compared to those in the placebo group. The between-intervention-group differences (treatment effects) in CV were 1.60 m/s (95% CI: 0.70, 2.40) for the median nerve and 2.10 m/s (95% CI: 1.50, 2.90) for the sural nerve. A significant difference in peak velocity (PV) was also observed in the sural nerve (2.10 m/s; 95% CI: 1.00, 3.20) after 12 months. Significant improvements in CV were only observed up to 6 months in the tibial motor nerve, 1.30 m/s (95% CI:

0.60, 2.20). There were no significant changes in serum biomarkers, transforming growth factor beta-1 (TGF-1), or vascular endothelial growth factor A (VEGF-A). After 6 months of washout, there were no significant differences from baseline between groups in nerve conduction parameters of all three nerves. Tocovid at 400 mg/day significantly improves tibial motor nerve CV up to 6 months, but median and sural sensory nerve CV in up to 12 months of supplementation. All improvements diminished after 6 months of washout.

JAMA Neurol 75(4):444-452, 2018 **IMPORTANCE** Management of painful diabetic peripheral neuropathy remains challenging. Most therapies provide symptomatic relief with varying degrees of efficacy. Tocotrienols have modulatory effects on the neuropathy pathway and may reduce neuropathic symptoms with their antioxidative and anti-inflammatory activities.
OBJECTIVE To evaluate the efficacy of oral mixed tocotrienols for patients with diabetic peripheral neuropathy.
DESIGN, SETTING, AND PARTICIPANTS The Vitamin E in Neuroprotection Study (VENUS) was a parallel, double-blind, placebo-controlled trial that recruited participants from January 30, 2011, to December 7, 2014, with 12 months of follow-up. This trial screened 14,289 patients with diabetes from 6 health clinics and ambulatory care units from 5 public hospitals in Malaysia. A total of 391 patients who reported neuropathic symptoms were further assessed with Total Symptom Score (TSS) and Neuropathy Impairment Score (NIS). Patients 20 years or older with a TSS of 3 or higher and an NIS of 2 or higher were recruited.
INTERVENTIONS Patients were randomized to receive 200mg of mixed tocotrienols twice daily or matching placebo for 12 months. Patients with hyperhomocysteinemia (homocysteine level >2.03 mg/L) received oral folic acid, 5mg once daily, and methylcobalamin, 500 μg thrice daily, in both groups.
MAIN OUTCOMES AND MEASURES The primary outcome was patient-reported neuropathy TSS (lancinating pain, burning pain, paresthesia, and asleep numbness) changes at 12 months. The secondary outcomes were NIS and sensory nerve conduction test result.
RESULTS Of 391 eligible patients, 300 were recruited (130 [43.3%] male; mean [SD] age, 57.6 [8.9] years; mean [SD] duration of diabetes, 11.4 [7.8] years) and 229 (76.3%) completed the trial. The TSS changes between the tocotrienols and placebo groups at 12 months (−0.30; 95%CI, −1.16 to 0.56; P = .49) were similar. No significant differences in NIS (0.60; 95%CI, −1.37 to 2.65; P = .53) and sensory nerve conduction test assessments were found between both groups. In post hoc subgroup analyses, tocotrienols reduced lancinating pain among patients with hemoglobin A1C levels greater than 8% (P = .03) and normohomocysteinemia (homocysteine level <2.03mg/L; P = .008) at 1 year. Serious adverse events in both groups were similar,

except more infections were observed in the tocotrienols group (6.7%vs 0.7%, *P* = .04). Results reported were of modified intention-to-treat analyses.
CONCLUSIONS AND RELEVANCE Supplementation of oral mixed tocotrienols, 400mg/d for 1 year, did not improve overall neuropathic symptoms. The preliminary observations on lancinating pain among subsets of patients require further exploration.

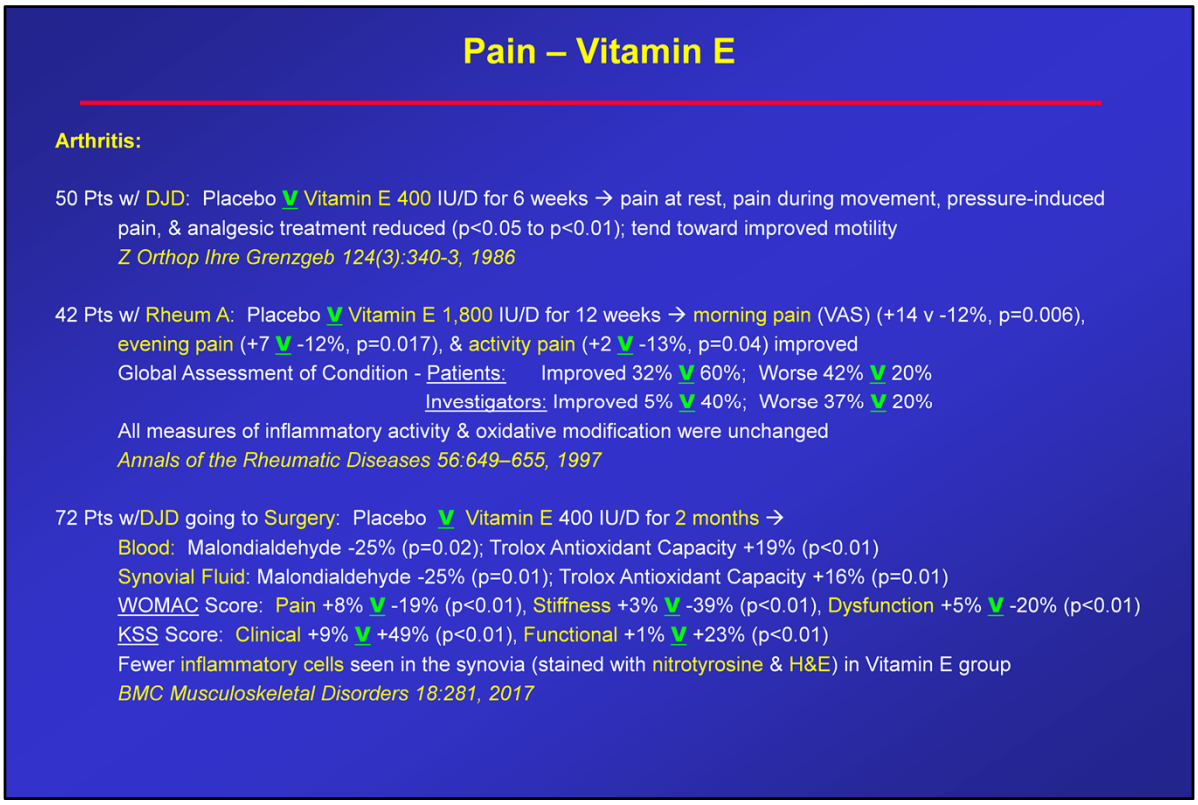
J Diabetes & Complications 19: 247– 253, 2005 **Abstract** **Background:** QR-333, a topical compound that contains quercetin, a flavonoid with aldose reductase inhibitor effects, ascorbyl palmitate, and vitamin D3, was formulated to decrease the oxidative stress that contributes to peripheral diabetic neuropathy and thus alleviate its symptoms. This proof-of-principle study assessed the efficacy and safety of QR-333 against placebo in a small cohort of patients with diabetic neuropathy.
Methods: This randomized, placebo-controlled, double-blind trial included 34 men and women (21–71 years of age) with Type 1 or 2 diabetes and diabetic neuropathy who applied QR-333 or placebo (2:1 ratio), three times daily for 4 weeks, to each foot where symptoms were experienced. Five-point scales were used to determine changes from baseline to endpoint in symptoms and quality of life (efficacy). Safety was assessed through concomitant medications, adverse events, laboratory evaluations, and physical examinations.
Results: QR-333 reduced the severity of numbness, jolting pain, and irritation from baseline values. Improvements were also seen in overall and specific quality-of-life measures. QR-333 was well tolerated. Eleven patients in the QR-333 group reported 23 adverse events (all mild or moderate); 4 in the placebo group reported 5 events (all moderate). One patient who applied QR-333 noted a pricking sensation twice, the only adverse event considered possibly related to study treatment.
Conclusions: From this preliminary safety study, it appears that QR-333 may safely offer relief of symptoms of diabetic neuropathy and improve quality of life. These findings warrant further investigation of this topical compound.

Clin J Pain 25(7):562-9, 2009 **Abstract** **Objectives:** Plasma vitamin C concentrations have been suggested to be related to pain modulation in post-herpetic neuralgia (PHN), an intractable neuropathic pain syndrome. In this study, we first compared plasma concentrations of vitamin C between healthy volunteers and PHN patients and then designed a symptom-based and mechanism-based approach to assess the analgesic effect of intravenous vitamin C on spontaneous and brush-evoked pain.
Methods: Study 1 was cross-sectional that enrolled 39 healthy volunteers and 38 PHN patients. Study 2 was a double-blinded, placebo-controlled intervention study, which comprised 41 patients randomly allocated into the ascorbate group and placebo. Each patient received normal saline infusion with or without ascorbate on days 1, 3,

and 5 and answered questionnaires that included side effects; numeric rating pain scale (NRS) on spontaneous and brush-evoked pain on days 1, 3, 5, and 7; and patient global impression of change on spontaneous and brush-evoked pain on day 7. [ascorbate (50 mg/kg body weight, maximum dose 2.5 g/d]

Results: Study 1 revealed that plasma concentrations of vitamin C were significantly lower in patients with PHN than in healthy volunteers ($P<0.001$). Study 2 showed that ascorbate treatment effectively restored plasma vitamin C concentrations in the patients and decreased spontaneous pain by 3.1 in NRS from baseline to day 7, as compared with a decrease of 0.85 in NRS by placebo treatment ($P<0.001$). Conversely, ascorbate treatment did not significantly affect brush-evoked pain. Ascorbate treatment also resulted in a better efficacy than placebo in patient global impression of change on spontaneous pain ($P<0.001$) on day 7 and did not affect brush-evoked pain. No side effects were observed.

Conclusions: Plasma vitamin C status plays a role in PHN, and intravenous ascorbate helps relieve spontaneous pain in PHN.



Z Orthop Ihre Grenzgeb 124(3):340-3, 1986 **Abstract** 50 patients with osteoarthritis were randomly assigned to two groups and treated over a period of 6 weeks with vitamin E-capsules (daily dose 400 I.E. d-alpha-tocopherylacetate) or an identical placebo preparation. The results of this double-blind controlled clinical trial showed that vitamin E was superior to placebo with respect to the relief of pain (pain at rest, pain during movement, pressure-induced pain) and the necessity of additional analgetic treatment (p less than 0.05 to p less than 0.01). Improvement of mobility was better in the group treated with vitamin E. However, this result was not statistically significant. The profile and the intensity of adverse reactions in both the vitamin E and placebo group was practically identical. This clinical study shows antiphlogistic efficacy of vitamin E in patients with osteoarthritis. In view of the possibility to reduce standard antiphlogistic, analgetic therapy together with the very good tolerance this result may be very important for the treatment of chronic rheumatic inflammatory disease.

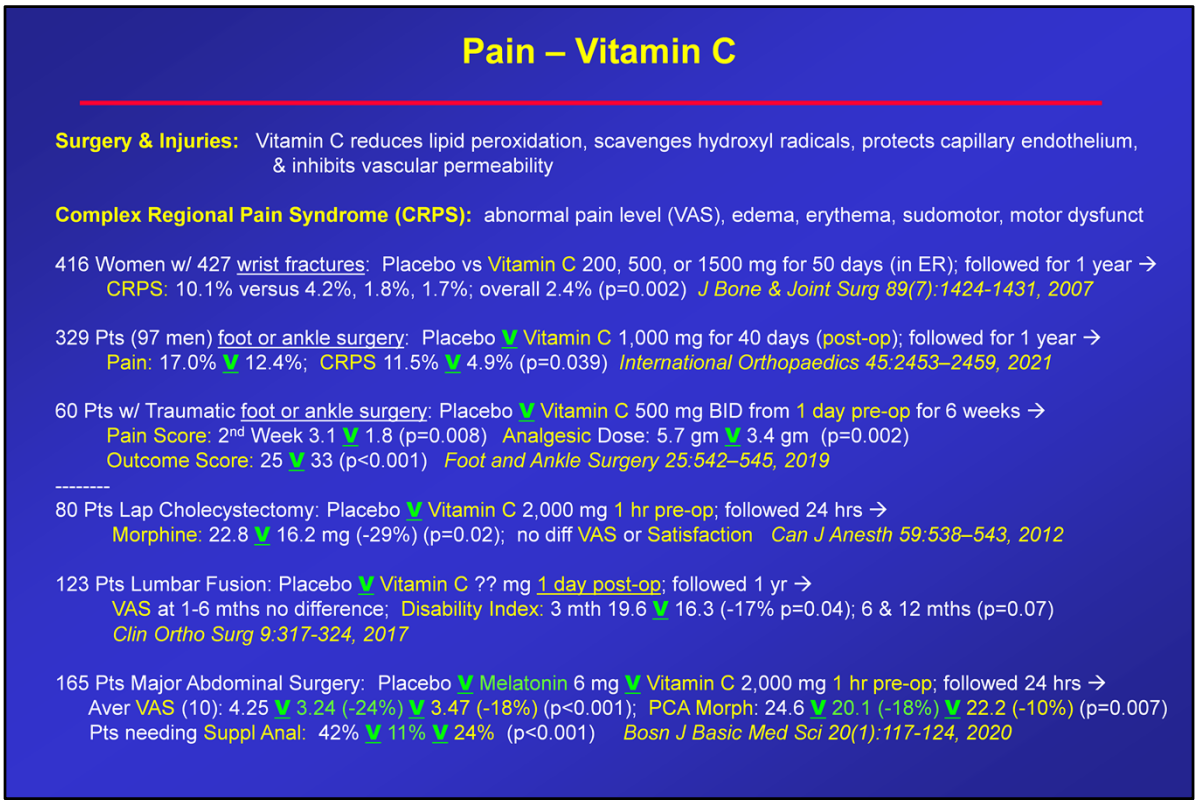
Annals of the Rheumatic Diseases 56:649–655, 1997 **Abstract** ***Objective*** Vitamin E, the most potent naturally occurring lipid soluble antioxidant has been suggested to possess both anti-inflammatory and analgesic activity in humans. This double blind and randomized study used a broad spectrum of clinical and laboratory parameters to investigate whether there was any additional anti-inflammatory or analgesic effects,

or both, of orally administered á-tocopherol in rheumatoid arthritis patients who were already receiving anti-rheumatic drugs.
Methods Forty two patients were enrolled and treated with á-tocopherol (n=20) at a dose of 600 mg twice a day (2x2 capsules) or with placebo (n=22) for 12 weeks. The following parameters were measured: (1) Three clinical indices of inflammation—the Ritchie articular index, the duration of morning stiffness, and the number of swollen joints; (2) three measures of pain—pain in the morning, pain in the evening, and pain after chosen activity; (3) haematological and biochemical measures of inflammatory activity; (4)
assays for the oxidative modification of proteins and lipids.
Results All laboratory measures of inflammatory activity and oxidative modification were unchanged. Furthermore, the clinical indices of inflammation were not influenced by the treatment. However, the pain parameters were significantly decreased after vitamin E treatment when compared with placebo.
Conclusion The results provide preliminary evidence that vitamin E may exert a small but significant analgesic activity independent of a peripheral anti-inflammatory effect, but which complements standard anti-inflammatory treatment.

BMC Musculoskeletal Disorders 18:281, 2017 **Abstract Background:** This study was performed to evaluate the antioxidative and anti-inflammatory effects of vitamin E on oxidative stress in the plasma, synovial fluid, and synovial tissue of patients with knee osteoarthritis.
Methods: Seventy-two patients with late-stage knee osteoarthritis scheduled for total knee arthroplasty were randomized to take oral placebo (Group A) or 400 IU of vitamin E (Group B) once a day for 2 months before undergoing surgery. The blood levels of endpoints indicating oxidative stress or antioxidant capacity, Knee Society Score (KSS), Western Ontario and McMaster Universities Osteoarthritis Index score (WOMAC), and adverse effects were compared before and after the intervention between the two groups. At surgery, these redox endpoints and histological findings were compared between the synovial fluid and synovial tissue.
Results: In blood samples, the pre-intervention of oxidative stress and anti-oxidative capacity were not different between Group A and Group B. In post-intervention blood samples, the Malondialdehyde (Group A 1.34 ± 0.10, Group B 1.00 ± 0.09, $p < 0.02$), Alpha tocopherol (Group A 15.92 ± 1.08, Group B 24.65 ± 1.47, $p < 0.01$) and Trolox equivalent antioxidant capacity (Group A 4.22 ± 0.10, Group B 5.04 ± 0.10, $p < 0.01$) were significantly different between Group A and Group B. In synovial fluid samples, the Malondialdehyde (Group A 1.42 ± 0.12, Group B 1.06 ± 1.08, p 0.01), Alphatocopherol (Group A 4.51, Group B 7.03, $p < 0.01$), Trolox equivalent antioxidant capacity
(Group A, 1.89 ± 0.06, Group B 2.19 ± 0.10) were significantly different between

Group A and Group B. The pre-intervention WOMAC score and KSS score were not different between Group A and Group B. The post-intervention WOMAC score was significantly improved in all categories in Group B (Pain: Group A 27.26 ± 0.89, Group B 19.19 ± 1.43, $p < 0.01$; Stiffness: Group A 8.23 ± 0.79, Group B 5.45 ± 0.73, p 0.01; Function: Group A 94.77 ± 4.22, Group B 72.74 ± 6.55, $p < 0.01$). The post-intervention KSS score was significantly improved in all categories in Group B (Clinical: Group A 25.31 ± 14.33, Group B 33.52 ± 16.96, $p < 0.01$; Functional: Group A 41.43 ± 16.11, Group B 51.61 ± 19.60, p 0.02). Significantly fewer synovial tissue cells were stained with nitrotyrosine and hematoxylin–eosin in Group B than in Group A. There were no differences in adverse effects or surgical complications between the groups.

Conclusion: Vitamin E is an effective antioxidant that can improve clinical symptoms and reduce oxidative stress conditions in patients with late-stage knee osteoarthritis.



J Bone & Joint Surg 89(7):1424-1431, 2007 **Abstract Background:** Complex regional pain syndrome type I is treated symptomatically. A protective effect of vitamin C (ascorbic acid) has been reported previously. A dose-response study was designed to evaluate its effect in patients with wrist fractures.

Methods: In a double-blind, prospective, multicenter trial, 416 patients with 427 wrist fractures were randomly allocated to treatment with placebo or treatment with 200, 500, or 1500 mg of vitamin C daily for fifty days. The effect of gender, age, fracture type, and cast-related complaints on the occurrence of complex regional pain syndrome was analyzed.

Results: Three hundred and seventeen patients with 328 fractures were randomized to receive vitamin C, and ninety-nine patients with ninety-nine fractures were randomized to receive a placebo. The prevalence of complex regional pain syndrome was 2.4% (eight of 328) in the vitamin C group and10.1% (ten of ninety-nine) in the placebo group (p = 0.002); all of the affected patients were elderly women. Analysis of the different doses of vitamin C showed that the prevalence of complex regional pain syndrome was 4.2% (four of ninety-six) in the 200-mg group (relative risk, 0.41; 95% confidence interval, 0.13 to 1.27), 1.8% (two of 114) in the 500-mg group (relative risk, 0.17; 95% confidence interval, 0.04 to 0.77), and 1.7% (two of 118) in the 1500-mg group (relative risk, 0.17; 95% confidence interval, 0.04 to 0.75). Early cast-related complaints predicted the development of complex regional pain syndrome (relative risk, 5.35; 95% confidence interval, 2.13 to 13.42).

Conclusions: Vitamin C reduces the prevalence of complex regional pain syndrome after wrist fractures. A daily dose of 500 mg for fifty days is recommended.

International Orthopaedics 45:2453–2459, 2021 **Abstract** **Purpose** Complex regional pain syndrome (CRPS) after foot and ankle surgery has a significant impact on the ability to walk. As the symptomatic treatment of this disaster complication is poor and has low efficacy, a preventive treatment would be beneficial. Vitamin C has been reported to be efficient in preventing CRPS in elective scheduled surgery. Few authors explored this efficiency in foot and ankle surgery. We, therefore, evaluated the efficacy of vitamin C in preventing this complication after foot and ankle surgeries for both trauma and elective surgery.
Material and methods Between January 2018 and December 2019, 329 patients were included in the study. We conducted a prospective randomized study on the efficiency of vitamin C (one group with and one without vitamin C) to prevent CRPS risk in patients operated in our institution on foot or ankle surgery. The incidence of CRPS after foot and ankle surgery was evaluated in both groups; the diagnostic of CRPS was made using the Budapest criteria associated with three-phase bone scintigraphy.
Results Among the 329 patients included in the study (232 women and 97 men), 121 patients were included in the vitamin C group and 208 in the control group (without vitamin C). Vitamin C was statistically linked with a decreased risk of CRPS (OR 0.19; CI 95% from 0.05 to 0.8; $p = 0.021$). Alcoholism and cast immobilization were increased risks factors of CRPS (respectively $p = 0.001$ and $p = 0.034$).
Conclusion Taking 1 g per day of vitamin C during 40 days after a foot or ankle surgery reduces the risk of CRPS.

Foot and Ankle Surgery 25:542–545, 2019 **Abstract** **Background:** Post-operative pain may adversely affect a patient's quality of life. Studies have shown that vitamin C, being an anti-oxidant and neuro-modulating agent, can help to reduce pain in a variety of clinical settings. The objective of this randomized controlled trial was to assess the effectiveness of vitamin C in reducing post-operative pain, analgesia requirements and improving functional outcome.
Methods: Patients with isolated foot and ankle trauma, who had undergone surgery, were randomly assigned to receive either vitamin C 500 mg or a placebo tablet twice a day. VAS score, analgesia requirement and functional outcome were assessed during their regular follow up. Results were compared and analyzed at the end of 3 months.
Results: The group which received vitamin C, showed improvement in VAS score at the end of second and sixth week of follow up, reduced analgesia requirements and improved functional outcome as compared to the placebo group.

Conclusions: This study shows that the supplementation of vitamin C in patients undergoing surgery for foot and ankle trauma helps to reduce analgesic requirements, improve VAS scores and achieve better functional outcome.

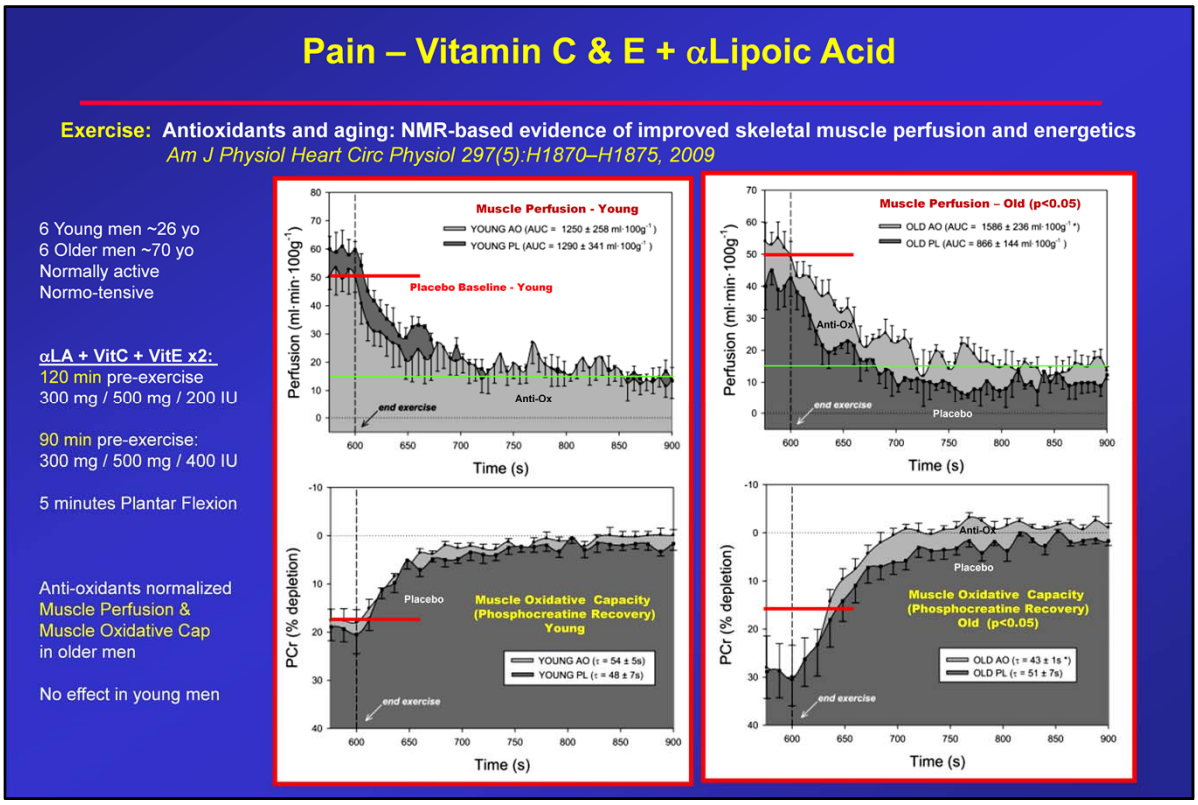
Can J Anesth 59:538–543, 2012 **Abstract Purpose** We designed a randomized double-blind placebo-controlled trial to assess the role of a single prophylactic dose of vitamin C (2 g) po in reducing the consumption of opioids postoperatively in patients undergoing laparoscopic cholecystectomy.
Methods Eighty adult patients were allocated to receive 2 g vitamin C po or placebo approximately one hour prior to induction of anesthesia. Following laparoscopic cholecystectomy, patients received morphine patient-controlled analgesia for 24 hr. The following data were assessed postoperatively in the post-anesthesia care unit at two, four, six, 12, and 24 hr: morphine consumption, verbal numerical rating scale scores for incisional pain and nausea/vomiting, and pruritus and sedation scores. The primary
outcome measure was 24-hr morphine consumption. Patient satisfaction was assessed before hospital discharge.
Results Morphine consumption was significantly lower in the vitamin C group vs the placebo group [16.2 (10.7) and 22.8 (13.8) mg, respectively; difference = 6.6 mg; 95% confidence interval, 1.1 to 12.1 mg; P = 0.02]. There was no difference in pain scores or side effects between the two groups. Satisfaction scores were similar in both groups.
Conclusion Our study showed that supplementation with vitamin C (2 g) po decreased morphine consumption in the postoperative period in patients undergoing laparoscopic
cholecystectomy.

Clin Ortho Surg 9:317-324, 2017 **ABSTRACT Background:** Vitamin C has critical features relevant to postoperative pain management and functional improvement; however, no study has yet evaluated the effectiveness of vitamin C on improving the surgical outcomes for spine pathologies. Thus, this study aimed to explore the impact of vitamin C on postoperative outcomes after single-level posterior lumbar interbody fusion (PLIF) for lumbar spinal stenosis in prospectively randomized design. We conducted a 1-year prospective, randomized, placebo-controlled, double-blind study to evaluate the impact of vitamin C on the postoperative outcomes after PLIF surgery.
Methods: A total of 123 eligible patients were randomly assigned to either group A (62 patients with vitamin C) or group B (61 patients with placebo). Patient follow-up was continued for at least 1 year after surgery. The primary outcome measure was pain

intensity in the lower back using a visual analogue scale. The secondary outcome measures were: (1) the clinical outcome assessed using the Oswestry Disability Index (ODI); (2) the fusion rate assessed using dynamic radiographs and computed tomography scans;
and (3) complications.
Results: Pain intensity in the lower back was significantly improved in both groups compared with preoperative pain intensity, but no significant difference was observed between the 2 groups over the follow-up period. The ODI score of group A at the third postoperative month was significantly higher than the score of group B. After the sixth postoperative month, the ODI score of group A was slightly higher than the score of group B; however, this difference was not significant. The fusion rates at 1 year after surgery and the complication rates were not significantly different between the 2 groups.
Conclusions: Postoperative pain intensity, the primary outcome measure, was not significantly different at 1 year after surgery between the 2 groups. However, vitamin C may be associated with improving functional status after PLIF surgery, especially during the first 3 postoperative months.

Bosn J Basic Med Sci 20(1):117-124, 2020 **ABSTRACT**

The analgesic benefit of melatonin and vitamin C as primary or adjuvant agents has been reported in various studies; however, their analgesic effects in the treatment of postoperative pain remain unclear. Thus, we aimed to evaluate the effect of single preoperative dose of oral melatonin or vitamin C administration on postoperative analgesia. In this study, we recruited 165 adult patients undergoing elective major abdominal surgery under general anesthesia. Patients were randomly divided into three equal (n = 55) groups. One hour before surgery, patients received orally melatonin (6 mg) in group M, vitamin C (2 g) in group C, or a placebo tablet in group P. Pain, sedation, patient satisfaction, total morphine consumption from a patient-controlled analgesia device, supplemental analgesic requirement, and the incidence of nausea and vomiting were recorded throughout 24 h after surgery. The mean pain score and total morphine consumption were found significantly lower in both M and C groups compared with group P ($p < 0.001$). There were no significant differences between group M and C with respect to pain scores ($p = 0.117$) and total morphine consumption ($p = 0.090$). Patients requested less supplemental analgesic and experienced less nausea and vomiting in groups M and C compared with group P.
In conclusion, preoperative oral administration of 6 mg melatonin or 2 g vitamin C led to a reduction in pain scores, total morphine consumption, supplemental analgesic requirement, and the incidence of nausea and vomiting compared with placebo.



Abstract *Am J Physiol Heart Circ Physiol 297(5):H1870–H1875, 2009*

We sought to examine the potential role of oxidative stress on skeletal muscle function with ad-vancing age. Nuclear magnetic resonance (NMR) was employed to simultaneously assess muscle perfusion (arterial spin labeling) and energetics (P NMR spectroscopy) in the lower leg of young (26 ± 5 yr, n = 6) and older (70 ± 5 yr, n = 6) healthy volunteers following the consumption of either placebo (PL) or an oral antioxidant (AO) cocktail (vitamins C and E and α-lipoic acid), previously documented to decrease plasma free radical concentration. NMR measurements were made during and after 5 min of moderate intensity (≈5 W) plantar flexion exercise.

AO administration significantly improved end-exercise perfusion (AO, 50 ± 5, and PL, 43 ± 4 ml·100 g·min) and post-exercise perfusion area under the curve (AO, 1,286 ± 236, and PL, 866 ± 144 ml/100 g) in older subjects, whereas AO administration did not alter hemodynamics in the young group. Concomitantly, muscle oxidative capacity (time constant of phosphocreatine recovery, τ) was improved following AO in the older (AO, 43 ± 1, and PL, 51 ± 7 s) but not the young (AO, 54 ± 5, and PL, 48 ± 7 s) group.

These findings support the concept that oxidative stress may be partially responsible for the age-related decline in skeletal muscle perfusion during physical activity and

re-veal a muscle metabolic reserve capacity in the elderly that is accessible under conditions of improved perfusion.

Pain

Effects of dietary supplements on PAIN:

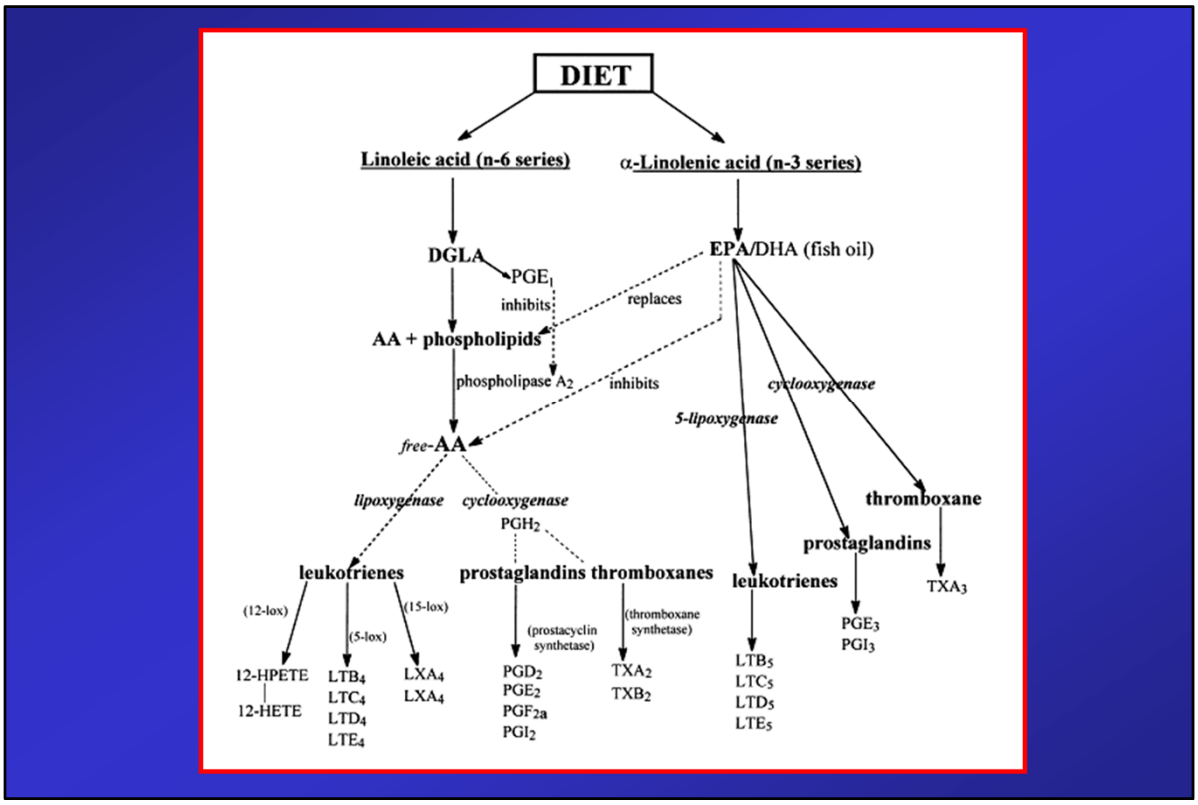
Omega-3 Fish Oil:
1. Maintains bone mass & cartilage integrity
2. Diabetic Neuropathy:
   - Improves cold & vibratory perception
   - Improves sensory scores & pain
3. Rheumatoid Arthritis:
   - Reduces the number of tender & swollen joints
   - Improves clinical scores
   - Patients reduce their anti-rheumatoid medications in 3-4 months
   - Reduces morning stiffness & fatigue
   - Improves daily activity
   - Improves patient's global perception of their disease activity
   - Reduces pain in 3-4 months
4. Degenerative Arthritis:
   - Prevents the development of pain & stiffness
   - Maintains physical function
   - Allows increased minutes of exercise
5. Promotes weight loss
6. Reduces post-operative pain
7. Exercise:
   - Reduces post-exercise soreness, muscle edema, reduced range of motion
   - Reduces post-exercise fatigue & pain
   - Improves muscle power
   - Reduces muscle inflammation
8. Covid-19: Reduces inflammation, pain, & fatigue
9. Migraines may be reduced
10. Symptoms in Parkinson Disease & early Dementia improve & progression slowed
11. Depression, anxiety, & ADHD probably improve

Alpha Lipoic Acid:
1. Diabetic Neuropathy: Improves nerve conduction, pain, & symptoms
2. Lumbo-Sacral Nerve Compression: Reduces pain & disability
3. Pain of Unknown Etiology (joint, nerve, or muscle): Reduced ~70% in 2 months

Vitamin E:
1. Diabetic Neuropathy: Improves nerve conduction, numbness, & freezing pain
2. Degenerative Arthritis:
   - Reduces pain at rest & with movement
   - Reduces stiffness & improves function
   - Reduces inflammation in joint
3. Rheumatoid Arthritis: Reduces morning pain & pain with exercise

Vitamin C:
1. Surgery:
   - Reduces post-operative pain
   - Reduces "Complex Regional Pain Syndrome"
   - Improves outcomes after joint & back surgery

Alpha Lipoic Acid + Vitamin E + Vitamin C:
1. Improves muscle blood flow after exercise in older men



Abstract *Int J Dev Neurosci Jul-Aug;18(4-5):383-99, 2000*
Linoleic and alpha-linolenic acid are essential for normal cellular function, and act as precursors for the synthesis of longer chained polyunsaturated fatty acids (PUFAs) such as arachidonic (AA), eicosapentaenoic (EPA) and docosahexaenoic acids (DHA), which have been shown to partake in numerous cellular functions affecting membrane fluidity, membrane enzyme activities and eicosanoid synthesis. The brain is particularly rich in PUFAs such as DHA, and changes in tissue membrane composition of these PUFAs reflect that of the dietary source. The decline in structural and functional integrity of this tissue appears to correlate with loss in membrane DHA concentrations. Arachidonic acid, also predominant in this tissue, is a major precursor for the synthesis of eicosanoids, that serve as intracellular or extracellular signals. With aging comes a likely increase in reactive oxygen species and hence a concomitant decline in membrane PUFA concentrations, and with it, cognitive impairment. Neurodegenerative disorders such as Parkinson's and Alzheimer's disease also appear to exhibit membrane loss of PUFAs. Thus it may be that an optimal diet with a balance of n-6 and n-3 fatty acids may help to delay their onset or reduce the insult to brain functions which these diseases elicit.

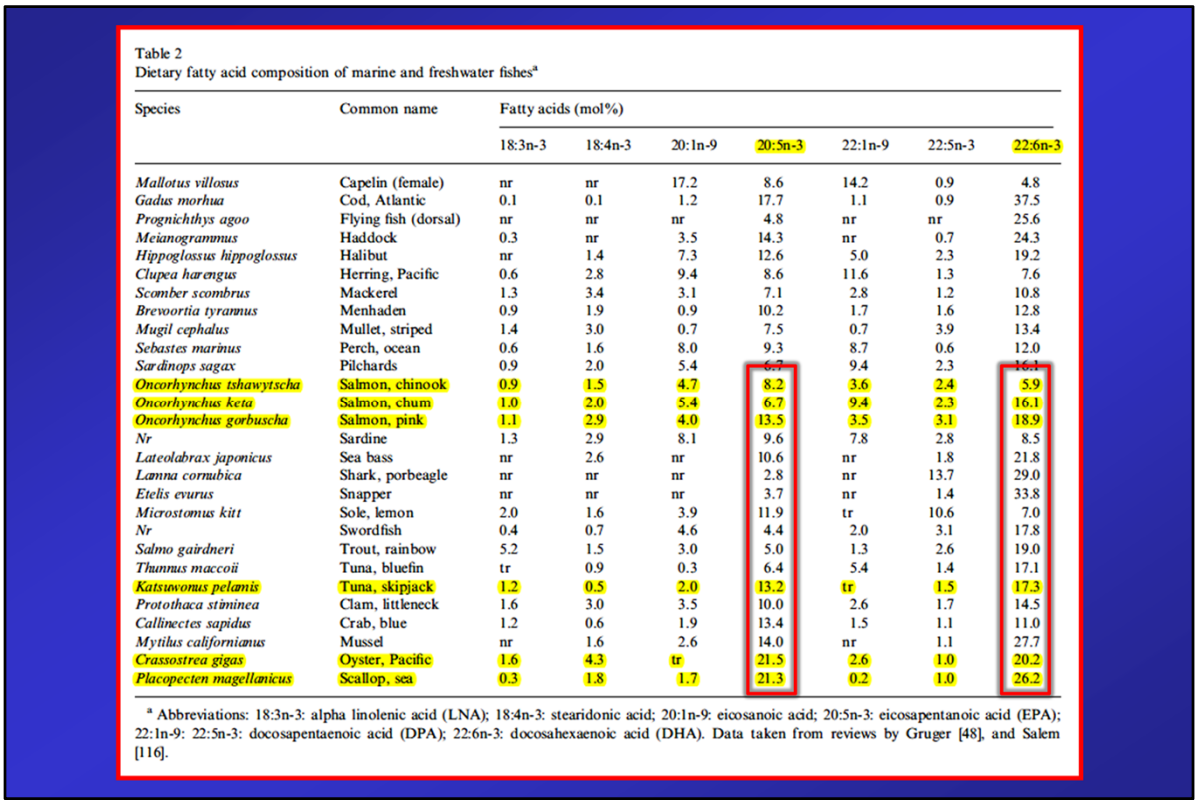


Table 2
Dietary fatty acid composition of marine and freshwater fishes[a]

Species	Common name	Fatty acids (mol%)						
		18:3n-3	18:4n-3	20:1n-9	20:5n-3	22:1n-9	22:5n-3	22:6n-3
Mallotus villosus	Capelin (female)	nr	nr	17.2	8.6	14.2	0.9	4.8
Gadus morhua	Cod, Atlantic	0.1	0.1	1.2	17.7	1.1	0.9	37.5
Prognichthys agoo	Flying fish (dorsal)	nr	nr	nr	4.8	nr	nr	25.6
Melanogrammus	Haddock	0.3	nr	3.5	14.3	nr	0.7	24.3
Hippoglossus hippoglossus	Halibut	nr	1.4	7.3	12.6	5.0	2.3	19.2
Clupea harengus	Herring, Pacific	0.6	2.8	9.4	8.6	11.6	1.3	7.6
Scomber scombrus	Mackerel	1.3	3.4	3.1	7.1	2.8	1.2	10.8
Brevoortia tyrannus	Menhaden	0.9	1.9	0.9	10.2	1.7	1.6	12.8
Mugil cephalus	Mullet, striped	1.4	3.0	0.7	7.5	0.7	3.9	13.4
Sebastes marinus	Perch, ocean	0.6	1.6	8.0	9.3	8.7	0.6	12.0
Sardinops sagax	Pilchards	0.9	2.0	5.4	6.7	9.4	2.3	16.1
Oncorhynchus tshawytscha	Salmon, chinook	0.9	1.5	4.7	8.2	3.6	2.4	5.9
Oncorhynchus keta	Salmon, chum	1.0	2.0	5.4	6.7	9.4	2.3	16.1
Oncorhynchus gorbuscha	Salmon, pink	1.1	2.9	4.0	13.5	3.5	3.1	18.9
Nr	Sardine	1.3	2.9	8.1	9.6	7.8	2.8	8.5
Lateolabrax japonicus	Sea bass	nr	2.6	nr	10.6	nr	1.8	21.8
Lamna cornubica	Shark, porbeagle	nr	nr	nr	2.8	nr	13.7	29.0
Etelis evurus	Snapper	nr	nr	nr	3.7	nr	1.4	33.8
Microstomus kitt	Sole, lemon	2.0	1.6	3.9	11.9	tr	10.6	7.0
Nr	Swordfish	0.4	0.7	4.6	4.4	2.0	3.1	17.8
Salmo gairdneri	Trout, rainbow	5.2	1.5	3.0	5.0	1.3	2.6	19.0
Thunnus maccoii	Tuna, bluefin	tr	0.9	0.3	6.4	5.4	1.4	17.1
Katsuwonus pelamis	Tuna, skipjack	1.2	0.5	2.0	13.2	tr	1.5	17.3
Protothaca stiminea	Clam, littleneck	1.6	3.0	3.5	10.0	2.6	1.7	14.5
Callinectes sapidus	Crab, blue	1.2	0.6	1.9	13.4	1.5	1.1	11.0
Mytilus californianus	Mussel	nr	1.6	2.6	14.0	nr	1.1	27.7
Crassostrea gigas	Oyster, Pacific	1.6	4.3	tr	21.5	2.6	1.0	20.2
Placopecten magellanicus	Scallop, sea	0.3	1.8	1.7	21.3	0.2	1.0	26.2

[a] Abbreviations: 18:3n-3: alpha linolenic acid (LNA); 18:4n-3: stearidonic acid; 20:1n-9: eicosanoic acid; 20:5n-3: eicosapentanoic acid (EPA); 22:1n-9: 22:5n-3: docosapentaenoic acid (DPA); 22:6n-3: docosahexaenoic acid (DHA). Data taken from reviews by Gruger [48], and Salem [116].

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