(N-3) Fatty Acids: Molecular Role and Clinical Uses in Psychiatric Disorders\textsuperscript{1,2}

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ABSTRACT

New discoveries in the field of neurophysiology and neuropharmacology have revealed the role of (n-3) fatty acids in controlling inflammation and protecting neuron cells from oxidative damage, preserving their function. It has also been thought that their psychoactive properties could be beneficial in certain psychiatric illnesses. This article discusses the newest discoveries of the affected activities by these fats in the cerebral cortex and the efforts that have been made to put them in practice in clinical trials in humans. In general, we were able to detect certain discord in the scientific community when designing placebo-based studies (mainly in establishing the appropriate therapeutic dose of (n-3) fatty acids, varying from the recommended dietary dose to an amount that may be 3 or 4 times higher), and in interpreting results. Although many studies have had the validity of their results questioned because of their small sample size, several studies seem to indicate that the (n-3) fatty acids are useful therapeutic tools in treating psychiatric conditions such as major depression, bipolar disorder, and several other disorders. Larger sample size studies are still required to better analyze the treatment potential of these agents. Adv. Nutr. 3: 257–265, 2012.

Introduction

(N-3) fatty acids are long-chained and unsaturated molecules only obtained by dietary intake of certain grains, such as flax-seed, canola, and walnuts, and sea fish (1). All varieties of (n-3) acids (mainly alpha-linoleic, DHA, and EPA) are essential components in mammalian metabolism, whether it be as anti-inflammatory molecules in the elongase-desaturase pathways that synthesize the different subtypes of (n-3) fatty acids (also directly inhibiting of the (n-6)–derived eicosanoids), inhibitors of excessive platelet activity, immune-modulating agents, and the main components in guaranteeing cell membrane stability (2–5). Unfortunately, as the scientific community finds, at an alarming rate, more and more evidence that these fats have cardioprotective, psychoactive, and cancer-fighting properties (6,7), the general population has decreased daily intake of (n-3)-rich foods, amounting to what has been generally called the Western diet, rich in sugars and with severe deficiencies in several micronutrients—vitamins, minerals, and (n-3) fatty acids (8,9). The western diet, compared with the (n-3)-rich foods of the Mediterranean diet, has been correlated with greater incidence of rectal cancer, cardiovascular diseases, and psychiatric illnesses (8).

It is widely accepted that the PUFA have an important role in many neural pathways and that their deficiency may be correlated with the occurrence of several psychiatric illnesses, such as major depression, bipolar disorder, obsessive-compulsive disorder, and anxiety disorders (10–14). Exploration of these mechanisms of action has inspired the pursuit of new treatment protocols that feature PUFA as an adjunctive or as a monotherapy for treatment of these diseases, with many surprising results.

Current status of knowledge

(N-3) fatty acids as membrane components

(N-3) fatty acids are responsible for almost 20% of the brain’s dry weight, and one third of all fats in the central nervous system belong to the PUFA class (15,16). In the neuron membrane, they are responsible for the maintenance of stability and conformity of receptors and structural ligands such as the Na\textsuperscript{+}/K\textsuperscript{+} ATPase, calcium, sodium and chloride ion channels, and caveolin proteins. Lack of these essential components can alter cell function in many ways. The molecular role of omega 3 fatty acids is synthetized in Figure 1.

Neuron membrane potential depends on an even flow of Na\textsuperscript{+} cations (through Na\textsuperscript{+} channels), which promotes depolarization and rapid repolarization of the neuron, Fig. 1 by the protein Na\textsuperscript{+}/K\textsuperscript{+} ATPase. This electric current is responsible for the release of neurotransmitters in the synaptic cleft.
and signal transmission throughout the cortex. Any malfunction in this system can lead to neuron hypofunctioning, slower responses, and decreased cognitive and limbic function (17–20).

Release of neurotransmitters from their intracytoplasmic vesicles is dependent on the docking of Ca\(^{2+}\) ions through vesicle-associated membrane proteins (21). Difficulty in ion transport, such as caused by membrane instability, can make the signal transmission more difficult in the cleft, therefore causing decreased neuron function in the central nervous system.

Proton leak is an essential process in which the neuron expends ~20% of its energy and is essential for the normal aerobic respiratory activity in these cells. This process depends on the integrity of membrane proteins in the mitochondria, which are, in their turn, dependent on the dietary intake of essential fats (21–24).

GABAergic function, promoted by a class of chloride receptors, is important to decrease the firing rate of other neuron systems, thus not permitting them to hyperfunction and to cause neuron damage. Its malfunction is also correlated with the occurrence of certain anxiety disorders such as generalized anxiety disorder. (N-3) fatty acid depletion can alter the conformation of the chloride ion channels, which may derail GABAergic receptor activity (25).

Caveolae are a special class of proteins that can help dock or separate certain membrane proteins and promote signal exchange between the nucleus and the cytosolic environment, increasing receptor exchange between both compartments, preventing membrane protein senescence and loss of function, and increasing gene transcription of membrane receptors (25,26). Several studies show their enforcing role in the signal transduction via the type 2 serotonin membrane receptor, enabling serotonergic activity in the prefrontal, parietal, and somatosensory cortex, having a protective effect against depression (27–29). Their promotion of glutamatergic activity via the L-type Ca\(^{2+}\) channel/glutamate receptor subunit N-methyl-D-aspartate A2B/voltage-dependent anion channel signaling is also responsible for preventing neuron death (30,31) and preventing several pathological processes that may lead to major depression, attention-deficit disorder, and dementia (31). Their action against dopamine receptors type D1 has proved to be useful in preventing the negative symptoms of schizophrenia (32).

In sum, the normal activity of the proteins involved in cell metabolism (transporters in the cell membrane and the mitochondria, voltage-dependent anion channels, Na\(^+\)/K\(^+\) ATPase) is intrinsically connected to the (n-3) fatty acid composition of the cell membrane, and any decrease in the concentration of these fats can lead to neuron hypofunctioning, serotonin depletion, increased neuron death, and decreased energy metabolism in the neurons (22–24), which may be responsible for causing more pernicious diseases such as dementia.
(N-3) fatty acids in cytosolic pathways
The physiological balance between concentrations of (n-6) and (n-6) fatty acids maintains lower synthesis of proinflammatory cytokines (33–37) (mainly IL-1β and TNF-α), lower synthesis of prostaglandins, leukotrienes, and phosphodiesterase type 4 (38,39). When there is low concentration of the (n-3) PUFA, the increase in tissue inflammation can lead to cAMP cleavage, lower levels of cAMP response element binding factor, and brain-derived neurotrophic factor, which reduces synaptic plasticity and neurotransmission and increases neuron damage and death (40).

The (n-3) PUFA are also responsible for interacting with several nuclear transcription factor receptors such as sterol regulatory element binding protein 1c, PPAR receptor type α, retinoic acid receptors, and retinoid X receptor, all of which have important roles in memory, cognition, and problem-solving (41,42). Recent studies have also indicated the role of (n-3) fatty acids in controlling RNA transcription of fatty acid synthase, an important regulator of lipid metabolism in the neuron, and cyclooxygenase-2, an enzyme necessary in the inflammation cascades (42). The discovery of these pathways have shed light to certain pathological processes that occur in the brain, such as the decrease of brain-derived neurotrophic factor production and activity in major depressive disorder (40) and the decreased production of nuclear binding elements in age-related cognitive decline (42,43) and dementia (44).

(N-3) fatty acid deficiency, inflammation, and the etiology of mental illness
It has been demonstrated that inflammation may be key to the development of mental illnesses by causing increased production of cytokines, migration of inflammatory cells, and activation of glial cells, mainly astrocytes (45–47). It is believed that this continuous inflammation can lead to neuron malfunction and eventually to neuron death because of the increase in oxidative stress in the brain (47) via several molecular pathways that include, but are not limited to, glutathione peroxidase, glial fibrillary acidic protein, and TNF-α/IL-18 (46–48). This damage may be reflected in decreased neurogenesis in the hippocampus, causing several symptoms correlating with mood disorders (48), such as major depression and obsessive-compulsive disorder, and when conducted for a long period, long-term and irreversible effects can be observed in the prefrontal cortex and dopaminergic nuclei, causing dementia and Parkinson’s disease (49).

The lack of (n-3) fatty acids can lead to neuron malfunction and death by other means, whether by affecting the energy metabolism of the neuron (17) or mRNA translation of important components for the cell—second messengers, transduction receptors, and survival signals (41–43). Other membrane transporters may be affected by the lack of these essential fats, which are involved in maintaining hydroelectrolytic balance in the neuron, and the continuous exchange of substrates and products of cytosolic metabolism (26). It has been shown that (n-3) fatty acid depletion can decrease the rate of production of neurotransmitters in the monoaminergic pathways and decrease the rate of neuron firing—in a process similar to that observed in depressed patients—with decreased production of serotonin and decreased activity in serotoninergic pathways (49).

Major depression
Rationale. There is solid research highlighting the importance of the (n-3) fatty acids in maintaining membrane integrity for the transport of tryptophan, the precursor of serotonin in the raphe nucleus, the maintenance of serotonin type 2 receptors in the prefrontal cortex, which are responsible for mood state, and the control of inflammatory and oxidative damage to serotoninergic neurons. Maes et al. (50) verified that there is abnormal (n-3) fatty acid metabolism in depressed patients, with rapid increase in monounsaturated fatty acids and inflammatory response, and that these alterations were not reversed in traditional antidepressant therapy in some volunteers. Peet et al. (51) determined the red blood cell membrane concentration of (n-3) fatty acids in depressed patients compared with controls. These measurements are thought to relate directly to the neuron membrane concentration of these same PUFA. This study revealed that not only are (n-3) fatty acid concentrations lower in depressed patients, but that this correlated with greater inflammatory activity, matched only when the control group samples were immersed in a peroxide solution.

Studies. Major depression has been considered a successful example of a treatable disease either by monotherapy or combination of these fatty acids with traditional selective serotonin uptake inhibitors. Nemets et al. (52) designed a placebo-controlled study of subjects already undergoing antidepressant therapy and found better outcome results in the treatment group with ethyl-EPA (E-EPA), a modified component of (n-3) fatty acid formulations. Jazayeri et al. (53) studied a group of 60 outpatients, divided in 3 subgroups: 1 group received fluoxetine 20 mg/d, the second group received EPA at a dose of 1 g/d, and the third received a combination of these 2 treatments. The result was a similar improvement in general symptoms of the monotherapy groups compared with baseline and a significantly greater improvement in the combined therapy group. Peet et al. (54) conducted a study that showed improvement in the patients treated with E-EPA, and who were refractory to selective serotonin uptake inhibitor monotherapy for major depression. Other clinical trials have found positive correlations between an increase in (n-3) fatty acid concentrations and improvement in symptoms, whether in major isolated depression (54–57), pediatric unipolar depression (58), and depression associated with Parkinson’s disease (59), with menopausal transition (60), and with aging in women (61).

However, certain trials could not find any correlation between administration of fatty acid doses and improvement of symptoms. Even though well conducted, there should be certain reservations when interpreting their results,
mainly because of disagreement in the literature concerning the appropriate therapeutic daily dose of these substances, and the appropriate DHA:EPA ratio that should be prescribed for these patients (56–62), making administrations unique. One example is Rogers et al. (63), who could not find any correlation between supplementation and improvement, although there were reservations about their use of oleic acids as an appropriate placebo, and their preparations with low concentrations of EPA, which is thought to be the most active component in relieving mood symptoms.

Highlighting postpartum depression, there was a great division of opinion concerning the effectiveness of treatment. Four studies found no correlation (61–64) between supplementation and improvement, and two others found a positive correlation (65,66), not yet clarifying whether there is any benefit in this approach.

**Bipolar disorder**

**Rationale.** (N-3) fatty acids appear to be natural membrane stabilizers, stabilizing calcium voltage-dependent channels, neuron firing, and inflammatory processes that possibly damage this cell layer (17–21,71). Noaghiul and Hibbeln (72), compiling epidemiological data from other studies on the prevalence of bipolar disorder (types I and II) found that the occurrence of these diseases was inversely correlated with high seafood consumption. Biochemical analysis of different age sectors in U.S. and European populations established the correlation between low concentration of (n-3) fatty acids and the higher prevalence of mood disorders and mood instability (73–75).

**Studies.** Studies on pediatric patients have shown promising results, although they are limited due to being open label and with a limited sample size (76–80). Fragou (81) conducted a placebo-controlled study in adults using 1–2 g/d of E-EPA and observed a modest improvement in severity of illness. This same author observed an increase in the concentration of N-acetylaspartate in supplemented patients, a substance with proven neurotrophic properties (75).

Other authors have shown that supplementation with EPA is both well tolerated and effective, although its use is considered optimal in adult patients with moderately intense symptoms. Results of a study revealed that even with doses almost 3 times the dietary recommendation (6 g/d), patients with severe symptoms or rapid cycling disorder did not present any improvement after treatment compared with placebo (85).

**Dementia**

**Rationale.** There is a clear link between the aging brain, cognitive decline, and decrease in the total concentration of DHA, EPA, and lipid markers (86). Long-term potentiation, an activity that permits the conversion of short-time memory into long-term memory, is also shown to be a process closely linked to DHA levels in the hippocampus (87). This potentiation is deficient in certain conditions such as Alzheimer’s disease, and supplementation with DHA in selected animals was able to restore neuron function to normal. DHA supplementation was also able to reduce concentration of β-amyloid protein, plaque burden, and tau protein, which are thought to be disease activity markers in Alzheimer’s disease (82,83,88,89).

**Clinical trials**

Studies are divided in their findings of the effectiveness of essential fatty acids in improving symptoms. Although some clinical trials have shown that age-related cognitive decline can be partially reversed by supplementation with DHA and EPA and that patients with organic brain damage or Alzheimer’s disease can have improvement in immediate and delayed memory function (90,91), others found no effect of supplementation (92–95). Other studies found mixed results, with improvement only in a cognitively impaired population and not in Alzheimer’s patients (96), in depressive symptoms associated with this disease (97), or only improving cognitive function in very mildly symptomatic patients (98).

**Attention-deficit hyperactivity disorder**

**Rationale.** There are no clear mechanisms that involve the role of essential fatty acid deficiency in generating hyperactive symptoms, although it can be inferred that their role as anti-inflammatory molecules and as membrane-stabilizing components may be responsible for such an effect (100–103). Population studies revealed that children with attention-deficit hyperactivity disorder had lower serum concentrations of PUFA, particularly EPA (100,102), and had a higher rate of oxidation of (n-3) fatty acids, which results in a lower serum concentration of these fats (103).

**Clinical trials**

Generally, trials were successful in confirming (n-3) fatty acid supplementation as feasible in reducing hyperactive symptoms (104–107), improving student-teacher relations, impulsive action (105), visual memory acquisition (106), and learning performance as a whole (107). There were certain exceptions to these studies (108,109), in which no effects were found.

**Schizophrenia and first-episode psychosis**

**Rationale.** First-episode psychotic patients show a change in the composition of N-acetylaspartate in the brain (110), with a rapid decrease in concentration of this substance. This is also correlated with development of more serious and lasting schizophrenic symptoms, greater neuron death, and decrease in dopaminergic function (111). There are studies that show that N-acetylaspartate is linked to neuron membrane integrity, and its concentrations are dependent on (n-3) fatty acid serum concentrations (110).

**Clinical trials**

Supplementation with E-EPA reduced markers for neuron death in first-episode psychosis (112), mainly glutathione, and the tolerability of neuroleptic medication in patients (113), also showing improvement in negative symptoms of
schizophrenia and neuroprotective effects in hippocampal neurons (114). Another study, conducted by Amminger et al. (115), showed that in a sample of 81 individuals with an ultrahigh risk of the development of a psychotic disorder, there was prevention of the development of negative symptoms and slowing of the progression to full schizophrenia in the intervention group with 1.2 g/d for a 12-wk period, followed by a 40-wk observation period compared with placebo. There was also effectiveness in reducing positive and negative symptoms in schizophrenic patients, whether in monotherapy or in association with neuroleptics, even when used in low doses (2 g/d) for brief periods of intervention (12 wk) (116–118). Only 1 study did not find any improvement when (n-3) fatty acids were added to the treatment (119).

**Autism**  
**Rationale.** There are no described molecular mechanisms that are affected by (n-3) fatty acids in this disorder, and the evidence that any nutritional or pharmacological component can produce any improvement is derived from a group of pilot studies (120–122).

**Clinical trials**  
Amminger et al. (123) conducted a pilot trial with 13 individuals with a diagnosis of autism for 6 wk in which 1.5 g total (n-3) fatty acid composition (7 g of DHA and 0.84 g of EPA) were given to the intervention group. They observed a statistically significant change in hyperactivity and stereotypy of behavior in individuals compared with the placebo group. Both Bent et al. (124) and Politi et al. (125) conducted similar placebo-controlled studies with autistic patients in childhood and adulthood, respectively, yet neither group could find statistically significant differences between groups. They observed that there was a tendency toward a small improvement of symptoms in the intervention group, however, which justified conducting studies with larger samples of individuals.

**Anorexia nervosa**  
**Rationale.** Two studies with female patients with anorexia revealed that these patients had an alteration in the EPA/arachidonic acid profile, a decrease in all fractions of (n-3) fatty acids, and a decrease in liver protein metabolism (126,127). It is thought that this disease has certain similarities to obsessive-compulsive disorder, with malfunction of serotonin pathways in the limbic system and prefrontal cortex and that the PUFA act in the same way as in depressive disorders, as an anti-inflammatory molecule, preventing neuron damage, stabilizing neuron membrane, and signal transduction throughout the affected pathways (3,5,12).

**Clinical trials**  
Only 2 trials have been conducted that used (n-3) fatty acids as therapeutic tools, in 1 trial as the only intervention and in another trial as an adjunctive therapy with fluoxetine. Ayton et al. (128) worked with only 7 patients, all of whom showed significant improvement in anxiety symptoms, yet there was no control group with which to make the necessary comparisons. Barbarich et al. (129) could not find any correlation between improvements in weight gain in the intervention group compared with PUFA placebo.

**Obsessive-compulsive disorder**  
**Rationale.** The change in membrane permeability and neuron activity in serotonergic pathways and increased inflammatory activity in the brain are thought to be a connection between a deficiency of essential fatty acids and the development of compulsive symptoms (46).

**Clinical trials**  
Only 1 clinical trial was conducted with a placebo-controlled crossover model for 6 wk for EPA, in which no correlation was found between intervention and improvement in symptoms (130).

**Borderline personality disorder**  
**Rationale.** Neural mechanisms have not yet been discovered in this psychiatric condition. There is 1 study that revealed a correlation between DHA supplementation and a decrease in aggression in otherwise healthy young adults (131). Sudden and usually unmotivated aggression is one of the main symptoms of bipolar disorder, and a similar improvement was thought to occur in these patients.

**Clinical trials**  
Only 1 trial concerning this disorder was conducted by Zanarini et al. (132). In it, 30 female subjects were divided in a 2:1 ratio for EPA/placebo treatment, for an 8-wk period. There were significant improvements in aggressive symptoms and depression scores; therapy was well tolerated and presented no serious side effects.

**Drug dependence**  
**Rationale.** Drug-dependent patients go through a period of great anxiety and individual psychological suffering, with combined physical symptoms that include, but are not limited to, tremors and pain. It is the phase of most relapses in drug users and is commonly referred to as “craving.” New theories clarify that the role of proinflammatory cytokines is indeed important in the establishment of this syndrome and that the EPA fraction of essential fats counteracts the toxic effects of these substances in the brain (133–135). The neuroprotective effect of (n-3) fatty acids in serotonin production and action in the prefrontal cortex may also be responsible for the preservation of the ability of planning and task execution, both of which are impaired during withdrawal and the establishment of craving symptoms.

**Clinical trials**  
Only 2 trials have tested the effects of PUFA in drug abusers, both conducted by Buydens-Branchey et al. (135, 136). In 2006, 1 trial tested anxiety symptoms in substance abusers before and after supplementation with an EPA+DHA.
mixture, total 3 g PUFA/d for 3 mo (135). The improvement in the supplementation group was statistically different compared with placebo patients, and even after discontinuing therapy, the intervention group maintained a lower score of anxiety symptoms than the placebo group after 3 mo (135). The other study, a pilot, included analyses of anger and anxiety symptoms in drug abusers and found that the (n-3) fatty acids group showed a greater improvement compared with placebo (136).

Conclusions
Most evidence suggests that (n-3) fatty acids act on many mechanisms involved in the physiopathology of mental illnesses. However, it is impossible, at the moment, to state emphatically whether the deficiency of essential fatty acids is the cause of these problems themselves or whether these molecules simply can counterbalance the effect of other causes of mental disorders.

The essential fatty acids, according to the knowledge gathered so far, present themselves as new viable agents in psychiatric treatment because they are generally well tolerated, with minimal or no side effects. The studies that have proved a positive correlation between improvement in symptoms and the intake of (n-3) fatty acids are validity. However, further studies are necessary to establish whether they are viable in larger and more complex placebo-controlled studies because the previous work done with (n-3) fatty acids lacked standardization of therapeutic doses and were generally conducted with small samples.

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Literature Cited
26. Head BP, Patel HH, Tsutsumi YM, Hu Y, Mejia T, Mora RC, Insel PA, Roth DM, Drummond JC, Patel PM. Caveolin-1 expression is
64. Mischoulon D, Papakostas GI, Dording CM, Farabaugh AH, Sonawalla SB, Agoston AM, Petrillo LF, Pascuillo E, Economou NI, Joffe H, et al. Omega-3 fatty acids 264


87. Martin DS, Spencer P, Horrobin DF, Lynch MA. Long-term potentiation in aged rats is restored when the age-related decrease in polyunsaturated fatty acid concentration is reversed. Prostaglandins Leukot Essent Fatty Acids. 2002;67:121–30.


