REVISIÓN

Docosahexaenoic acid (DHA), an essential fatty acid for the proper functioning of neuronal cells: their role in mood disorders

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RESUMEN

El acido docosahexaenoico (DHA), un acido graso esencial para la adecuada función de las células neuronales: su efecto en los desordenes del comportamiento.

El cerebro y el sistema nervioso son tejidos con un alto contenido de dos ácidos grasos poliinsaturados: el ácido araquidónico (20:4, omega-6, AA) y el ácido docosahexaenoico (22:6, omega-3, DHA). A pesar de la abundancia de estos ácidos grasos en dichos tejidos los mamíferos no los pueden sintetizar de novo. Sin embargo, la concentración de estos ácidos grasos puede ser modificada por la dieta. El AA y el DHA pueden ser aportados por la dieta como tales (preformados) o a partir de los respectivos precursores de origen vegetal. El ácido linoleico, precursor del AA es muy abundante en la dieta occidental, por lo cual la formación de AA no es restrictiva. Por el contrario, el ácido alfa linolénico, cual es el precursor del DHA, es mucho menos disponible a partir de nuestra dieta, siendo muy restringido el consumo de este ácido graso en algunas poblaciones. El desarrollo del sistema nervioso ocurre de forma excepcionalmente rápida durante la última etapa del período gestacional y durante la primera etapa del período post natal. En estas etapas se requieren importantes cantidades de ácidos grasos poliinsaturados omega-6 y omega-3, particularmente de DHA, ya que estos ácidos grasos son críticos para el crecimiento neuronal y para el desarrollo y función del cerebro y la retina. En esta revisión se analizan varias funciones del DHA en el sistema nervioso, su metabolismo en los fosfolípidos y su función en diferentes desordenes neurológicos y del comportamiento, tales como la enfermedad de Alzheimer, diferentes formas de depresión, y otras.

PALABRAS–CLAVE: Acido docosahexaenoico – Desordenes del comportamiento – Fosfolípidos cerebrales – Función neuronal.

SUMMARY

Docosahexaenoic acid (DHA), an essential fatty acid for the proper functioning of neuronal cells: their role in mood disorders.

The brain and the nervous system are tissues with high contents of two polyunsaturated fatty acids: arachidonic acid (20:4, omega-6, AA) and docosahexaenoic acid (22:6, omega-3, DHA). Despite their abundance in these tissues, AA and DHA cannot be re-synthesized in mammals. However, the concentration of these fatty acids can be modulated by dietary intake. AA and DHA must be provided by the diet as such (preformed) or through the respective omega-6 and omega-3 precursors from vegetable origin. Linoleic acid, the precursor of AA is very abundant in the western diet and therefore the formation of AA from linoleic acid is not restrictive. On the other hand, alpha linolenic acid, the precursor of DHA is less available in our diet and preformed DHA is highly restrictive in some populations. During the last period of gestation and during the early post natal period, neurodevelopment occurs exceptionally quickly, and significant amounts of omega-6 and omega-3 polyunsaturated fatty acids, especially DHA, are critical to allow neurite outgrowth and the proper brain and retina development and function. In this review various functions of DHA in the nervous system, its metabolism into phospholipids, and its involvement in different neurological and mood disorders, such as Alzheimer's disease, depression, and others are revised.

KEY-WORDS: Brain phospholipids – Docosahexaenoic acid – Mood disorders – Neural function.

1. INTRODUCTION

During the last trimester of intrauterine foetal life and the first 2 years of childhood, the brain undergoes a period of rapid growth and development which has been defined as the "brain growth spurt" (McCann & Ames, 2005). Research over the last 30 years has established that long-chain omega-6 and omega-3 polyunsaturated fatty acids (LC-PUFAs) are critical for the development and functioning of the brain and nervous system and for the proper growth and development of newborns and children (Uauy & Valenzuela, 2000; Wainwright, 2002; Valenzuela *et al.*, 2006). Brain growth spurt is associated with a rapid increase in the incorporation of LC-PUFAs into phospholipids that form the brain cortex. The brain and nervous system, the retina and the sperm have the highest concentration of the omega-3 LC-PUFA docosahexaenoic acid (22:6, DHA), the most highly polyunsaturated fatty acid found in mammals. In the retina, a tissue, which from the phylogenetic point of view can be considered of neural origin (Jeffrey *et al.*, 2001), DHA, is concentrated in the outer segments of photoreceptors, in which nearly 50% of the fatty acid

is associated with the phospholipids of these cells (Fliesler & Anderson, 1983). In neurons, DHA is mainly associated with membrane phospholipids where they affect membrane fluidity and regulate the function of many functional membrane-associated proteins (Rapoport, 2001). DHA, when liberated from membrane phospholipids by specific phospholipases, may act as a regulatory molecule, as nuclear receptor ligand or can be converted into other signalling molecules, collectively known as docosanoids such as docosatrienes, and the tri-hydroxy-containing autacoids resolvins (Hong et al., 2003; Serhan & Chiang, 2004). This conversion occurs not in neuronal cells but in brain glial cells (Hong et al., 2003). The novel endogenous DHA-derived 10, 17 S-docosatriene, formed via a 15-lipooxygenase-like enzyme and identified as neuroprotectin D1, is a potent regulator of an intrinsic neuroprotective, anti-inflammatory and antiapoptotic gene-expression program that promotes survival in a stressed human brain (Lukiw et al., 2005; Lukiw & Bazan, 2006).

It is well known that a nutritional deficiency in omega-6 essential fatty acids in humans may results in several pathological abnormalities such as reduced growth, skin lesion, fatty liver, and reproductive failure (Holman, 1998). However, neuronal functioning appears not to be affected by nutritional omega-6 fatty acid deficiency. On the other hand, a nutritional deficiency in omega-3 fatty acids results in several neuronal specific effects such as reduced learning capacity, memory impairment, abnormal electroretinogram, impaired vision and numbness (Connor et al., 1992; Benatti et al., 2004). The different pathological defects that occur with either omega-6 or omega-3 fatty acid deficiency demonstrated that these two types of fatty acids are not metabolically interchangeable; omega-6 cannot substitute omega-3 fatty acids or vice-versa. Mammals cannot transform omega-6 fatty acids into omega-3 fatty acids (Kang, 2005). Most of the experimental research about omega-6/omega-3 fatty acid deficiency has been focussed on the consequences of omega-3 deficiency, because omega-6 deficiency in humans is extremely rare. The western diet provides sufficient amounts of omega-6 fatty acids but very low amounts of omega-3 fatty acids (Simopoulos, 2002) because preformed DHA is scarce in some populations.

2. DHA IN THE BRAIN AND NERVOUS SYSTEM

Within neurons, DHA is highly concentrated in membrane phospholipids, mainly as phosphatidylethanolamine and phosphatidylserine, the latter being the major acidic phospholipid in brain cell membranes (Breckenridge et al., 1972). Phospholipids which make up about one quarter of the solid matter in the brain are also an integral part of the vascular system on which brain function and nutrition depend. DHA makes up 15-20% of the fatty acid composition of the brain cortex, and when incorporated into phospholipids may improve the efficiency of synaptic membrane vesicles in fusion events which are fundamental for neurotransmission (Salem et al., 2001). DHA may also function in synaptic signalling either as a free fatty acid, as a metabolite or incorporated into phospholipids. DHA is also highly concentrated in growth cones during neurite outgrowth where it may be important for maximal neurite growth during brain development which occurs mainly during the perinatal period (Martin & Bazan, 1992). In the adult, DHA is found in neuronal dendrites, where it may be involved in the extension and establishment of the dendritic arborization which occurs during memory formation and the acquisition of learning capacities, modifications which originate in brain plasticity. Additionally, DHA may be important for the efficient regeneration of axons and dendrites in some brain regions, such as the cerebellum and hippocampus, after neuronal injury. Supplementing cultured neuronal cell types with AA and DHA at low concentrations significantly increases neurite outgrowth in several neuronal cell types, principally those from hippocampus (Calderon & Kim, 2004). However, there is a limit to the amount of AA to be added because at higher concentrations this fatty acid is cytotoxic. DHA, however, shows stimulant effects and no cytotoxicity in a wide range of concentrations (Ikemoto et al., 1997).

2.1. The role of DHA in neuronal phospholipid synthesis

DHA appears to enhance neurite outgrowth by several mechanisms which include an increase in the synthesis of specific phospholipids (Calderon & Kim, 2004). In differentiating and mature neurons DHA is preferentially incorporated into phospholipids rather than into triglycerides. During the synthesis of neuronal phospholipids, DHA is acylated to the sn-2 position of phospholipids to generate phosphatidic acid, which is the precursor of phosphatidylinositol. However, most of the phosphatidic acid is subsequently dephosphorylated to generate diacylglycerol, which is further metabolized into phosphaditidylcholine, phosphatidylethanolamine, phosphatidylserine and triglycerides, all of these molecules containing DHA specifically at the sn-2 position (Calderon & Kim, 2004). Therefore, it appears that diacylglycerols containing DHA at the sn-2 position are preferentially transformed into phospholipids. This specific transformation occurs through the action of specific enzymes. For example, diacylglycerol molecules that contain DHA in the sn-2 position are the preferred substrate of enzyme ethanolamine phosphotransferase which converts diacylglycerol to phosphatidylethanolamine through the covalent linking of ethanolamine to the sn-3 position of diacylglycerol (Holub, 1978). Subsequently, phosphatidylethanolamine can be converted to phosphatidylserine through the exchange of the nitrogen base with free serine (Mozzi et al., 2003).

Every cell type typically maintains a constant ratio of phospholipid species and a change in one species of phospholipids into another may often alter the level of another species (Araki & Wurtman, 1998). In neurons, the bulk of DHA is incorporated into phosphatidylethanolamine and to a lesser extent to phosphatidylserine. On the other hand, the majority of AA is incorporated into phosphatidylinositol and phosphatidylethanolamine. The bulk of other unsaturated fatty acids, such as oleic acid, is incorporated mainly into phosphatidylinositol. The differential incorporation of AA and DHA into phosphatidylinositol, phosphatidylserine and phosphatidylethanolamine, compared with phosphatidylcholine, may make an important contribution to their specific functions in neurons. The exact mode of action of DHA-containing phospholipids in brain functions is at present not known but there might be a relationship between their effect on the blood-brain barrier, membrane structure and fluidity in the activity of some specific enzymes, neural signalling, ionic channel permeability or the formation and control of nerve growth factors (Kitajka *et al.*, 2002). All these effects may be involved in the brain's cognitive functions such as memory and learning.

2.2. The role of DHA in membrane neuronal function

The quantity of double bonds in a fatty acid is directly related to the flexibility of the molecule. Saturated fatty acids, such as palmitic acid (16:0) or stearic acid (18:0), are rigid. This rigidity allows saturated fatty acids to pack together tightly and form a solid structure at lower temperatures. Phospholipids formed by these fatty acids are also rigid structures. The introduction of double bonds into a fatty acid causes a "kink" in its structure which modifies its spatial conformation. DHA, which has six double bonds, may adopt many countless conformations because the molecule can rotate around C-C bonds but not around the rigid six C = C that conform its high polyunsaturation (Feller *et al.*, 2002). The highly flexible structure of DHA will not allow phospholipids containing DHA to pack tightly together, resulting in a significant increase in membrane fluidity relative to phospholipids formed only by saturated fatty acids. Membranes having high contents of DHA may also increase the efficiency of membrane fusion events which are important in neurotransmission (Teague *et al.*, 2002). Additionally, an increased fluidity of membranes appears to be important for increasing the rate at which membrane protein-protein interaction occurs within the phospholipid's membrane bilayer. Fluidity is especially relevant in the outer segments of retinal photoreceptors where the activation of G type protein transducin by the rhodopsin-metarhodopsin interaction events occurs within the phospholipids of photoreceptor cells. This process does not occur efficiently when the level of DHA in the phospholipids of vision cells is reduced either during normal ageing or by pathological causes (Niu *et al.*, 2004). Mitochondrial phospholipids are also enriched in DHA. High DHA in mitochondrial membranes may increase the efficiency of the electron transport chain and the ADP-phosphorilation process by increasing the lateral movement of proteins within the membrane bilayer, thus facilitating protein-protein interactions (Valentine & Valentine, 2004). Additionally, there is a direct correlation between the DHA content of mitochondrial phospholipids and the permeability of the inner membrane to protons (Hulbert, 2003), thus improving the efficiency of energy production through oxidative phosphorilation. It is generally concluded that DHA positively influences mitochondrial energy function.

2.3. DHA and the activity of neuronal enzymes

Receptor functioning and the activation of proteins involved in neuronal signalling transduction can be influenced by DHA, either as a free fatty acid and/or when it is incorporated into membrane phospholipids. DHA is concentrated in the phospholipids of neural tissues, including the hippocampus (Ahmad *et al.*, 2002) which is involved in learning as well as in memory storage. Recently, we demonstrated that DHA supplementation to mother rats during the perinatal period, increases the DHA content of different brain segments of the pups, including hippocampus, and improves the learning and memory capacities of the pups when evaluated through the Skinner box test (Valenzuela *et al.*, 2008). As part of the diacylglycerol molecule, DHA enhances the diacylglycerol-dependent activation of the protein kinase C (PKC) (Chen & Murakami, 1994). It is interesting that PKC has an essential requirement for phosphatidylserine (Nishizuka, 1995), which contains a high concentration of DHA. However, in vitro evidence is suggesting that unesterified DHA may competitively inhibit phosphatidylserine dependent PKC activation. Unesterified AA either stimulates or has no effect on PKC activity (Seung Kim *et al.*, 2001), showing that activation of the enzyme by omega-3 fatty acids may be specific to these fatty acids. Another example of a protein whose function is modified by DHA is Na+, K+ ATPase, also known as sodium pump, which is an integral protein of the neuronal membrane found in higher concentration at the axonal nodes of Ranvier. The primary neuronal function of this ATPase is to generate and maintain Na+ and K+ gradients necessary to maintain the resting potential of neuronal membrane. The activity of Na+-K+ ATPase is increased in the sciatic nerve of rats that are supplemented with DHA (Gerbi *et al.*, 1998).

2.4. Inhibition of neuronal apoptosis by DHA

Neuronal cell survival is highly dependent on the presence of trophic nerve factors which influence downstream signalling pathways. Modifications in the

concentration and/or number of these factors may lead to apoptotic cell death. Early signs of apoptosis include the loss of intracellular water, an increase in cytoplasmic calcium concentration, the releasing of cytochrome c from mitochondria and the translocation of phosphatidylserine to the outer leaflet of the plasma membrane (Sastry & Rao, 2000). The activation of the caspase-3 enzyme by self-cleavage results in the death of cells by apoptosis (Nagata, 1997). The prevention of apoptosis by DHA incorporation into phospholipids has been reported for rat retinal photoreceptors (Rotstein *et al.*, 1997), HL-60 cells (Kishida *et al.*, 998), and Neuro 2A cells (Kim *et al.*, 2000). Additionally, an increased dietary intake of DHA prevents apoptosis in mouse retinal photoreceptors when subjected to N-methyl-N-nitrosourea, a potent inducer of apoptosis (Moriguchi *et al.*, 2003). DHA accumulation in phospholipids, mainly in phosphatidylserine, appears to promote neuronal survival under adverse conditions (Kim *et al.*, 2000).

As discussed above, in the nervous system, DHA is incorporated primarily into anionic phospholipids such as phosphatidylserine and phosphatidylethanolamine (Aid *et al.*, 2003). Phosphatidylserine is synthesized from phosphatidylethanolamine or phosphatidylcholine by the serine replacement of ethanolamine or choline, respectively, in a base-exchange reaction. Phosphatidylserine is involved in a series of cell-signaling events. The supplementation of cells with unesterified DHA promotes phosphatidylserine biosynthesis (Kim & Hamilton, 2000). The enrichment of DHA in phosphatidylserine and its effect on phosphatidylserine biosynthesis are most likely due to the fact that phospholipid species containing DHA are the best substrates for phosphatidylserine synthesizing enzymes (Kim *et al.*, 2004). There is not a direct correlation between the level of phosphatidylserine and DHA content in different brain segments. The antiapoptotic effect of DHA in neurons occurs only when the fatty acid is added to cultured cells or when experimental animals have been treated previously with DHA, which may suggest that these effects are due to the incorporation of DHA into different phospholipids. It is interesting to note that in other non-neuronal cell types, DHA actually promotes apoptosis. For example, in CaCo-2 cells, a colon cancer cell line, DHA induces apoptosis by "downregulating" reducing the expression of antiapoptotic genes and increasing the expression of proapoptotic genes (Narayanan *et al.*, 2001). Therefore, the antiapoptotic effects of DHA-containing phosphatidylserine are probably specific to neuronal cells and critical for the long-term survival of these cells.

2.5. DHA and the regulation of gene expression in neurons

It has been demonstrated that polyunsaturated omega-3 fatty acids can modify gene expression by binding to specific receptors and transcription factors in the liver and adipose tissue. Receptors activated by DHA include retinoid X, peroxisome proliferator activated receptors (PPARs), hepatic nuclear receptor, and sterol regulatory element binding protein (SREBP) receptor (Mata de Urquiza *et al.*, 2000). The activation of each of these proteins modulates the expression of genes involved in the metabolism of glucose, fatty acid, triglyceride, and cholesterol. Of these proteins, the retinoid X receptor is present in significant levels in the brain, and DHA is an effective ligand and activator of the retinoid X receptor protein (Lengqvist *et al.*, 2004). Activation of gene expression by DHA is not restricted to brain cells, the fatty acid activates several genes in other tissues, like liver or adipose tissue (Kitajka *et al.*, 2002). In rat brain cells, the stimulation of peroxisomal proliferator activated receptor β (PPAR β) and resulted in the upregulation of the mRNA encoding a protein that converted DHA to the acyl-CoA derivative (Marszalek *et al.*, 2005). Upon alteration of the expression of genes involved in lipid metabolism, the optimal environment for neurite outgrowth can be achieved during neuronal differentiation and brain formation. For example, omega-6 and omega-3 PUFAs have been shown to decrease the expression and the activity of Δ-9 desaturase, the enzyme that converts estearic acid (18:0) to oleic acid (18:1, omega-9) (Sessler *et al.*, 1996). This effect may be important to ensure that saturated fatty acids, whether newly synthesized or taken in from the diet, are available for the insertion of phospholipids into the sn-1 position as they are synthesized. Several studies have demonstrated that the DHA increasing effect on neurite outgrowth may be, in part, a consequence of the DHA stimulation of the expression of genes that promote phospholipids synthesis (Barcelo-Coblijn, 2003; Puskas *et al.*, 2003). Using microarray gene expression methodology, it has been demonstrated that fish oil or DHA supplementation can modify the expression of many of the genes of the brain and retina involved in signal transduction, eicosanoid production, synaptic plasticity, and energy metabolism in rats (Rojas *et al.*, 2003).

3. DHA AND ALTERATIONS OF NEURONAL FUNCTIONING AND MOOD DISORDERS

During periods of nutritional deficiency of omega-3 fatty acid, DHA is retained to depletion from the phospholipids of neurons through at least two different mechanisms. First, DHA released from membrane phospholipids is rapidly reacylated to specific phospholipids. Second, there is a significant reduction in the rate of transfer of DHA out of the nervous system through the blood-brain-barrier. Many neurodegenerative conditions, such as Alzheimer's disease, retinal affections, and some peroxisomal disorders (Zellweger syndrome and adrenoleucodisthrophy) are associated with

reduced levels of omega-3 fatty acids. Mood disorders, such as depression, schizophrenia, and post partum depression, have also been associated with modifications in DHA metabolism. Epidemiological, experimental and clinical data favour the hypothesis that DHA could play a role in the pathogenesis and eventually in the treatment of these diseases (Parker *et al.*, 2006; Stahl *et al.*, 2008).

3.1. Alzheimer's disease

Alzheimer's disease is a late-onset, progressive, neurodegenerative disease of heterogeneous origin which is devastating both to the afflicted person and to that person's family. The pathology is characterized by the formation of amyloid plaques, neurofibrillary tangles, and dystrophic neurites. Data from numerous epidemiological studies suggest an inverse correlation between DHA intake and the likelihood of developing Alzheimer's disease. A reduction in the level of total phospholipids as well as a decrease in DHA has been described in various cerebral areas in Alzheimer's disease patients (Prasad *et al.*, 1998). Studies have demonstrated that the levels of phosphatidylethanolamine, which is enriched in DHA, and of phosphatidylinositol, which is enriched in AA, are significantly reduced in the brain of individuals affected by Alzheimer's disease. Specifically, there is a significant reduction in the amount of DHA in the frontal cortex and hippocampus phospholipids of patients with Alzheimer's disesase (Soderberg *et al.*, 1991). The pretreatment of rats with DHA protected the animals against the memory loss which typically occurs when they are infused with Alzheimer disease Aβ peptide, which triggers a synapse loss (Coleman & Yao, 2003). DHA inhibits the accumulation of insoluble Aβ peptide, partially by decreasing cholesterol levels in the detergent insoluble membrane domains (rafts) of the cerebral cortex (Hashimoto *et al.*, 2005). It has been recently demonstrated that the effect of DHA in the reduction of insoluble Aβ peptide is attributable to a decrease in the steady-state levels of presenilin 1 (Green, *et al.*, 2007). In cognitive tests, animals expressing high levels of a mutant amyloid precursor protein, showed low levels of DHA in brain phospholipids. Additionally, the activity of phospholipase A2, which is involved in the liberation of AA from brain phospholipids, increases in the brain of patients with Alzheimer's disease, suggesting that an increased generation of AA-derived eicosanoids, which are antagonist of DHA-derived docosanoids, may contribute to the aetiology of Alzheimer's disease. It has been proposed that DHA-derived neuroprotectin induces an antiapoptotic and neuroprotective gene-expression program that regulates the secretion of Aβ peptide, resulting in the modulation of inflammatory signalling, neuronal survival, and the preservation of brain cell function (Lukiw *et al.*,

2005). The typical Western diet provides <30% of the 200-300 mg/day of DHA recommended by Expert Panels. Epidemiology shows a risk reduction of 60% associated with a modest increase in DHA intake or plasma levels. DHA works well in slowing down Alzheimer's disease pathogenesis in mice with a human Alzheimer's disease gene (Calon *et al.*, 2004). DHA provided by supplementation (e.g. fish meals, fish oil capsules, or other forms of DHA supplementation), could restore DHA deficiency in membrane phospholipids in the cerebral cortex of patients with Alzheimer's disease (Connor & Connor, 2007). DHA, together with natural antioxidants, may exert general anti-Alzheimer's and anti-aging benefits (Cole *et al.*, 2005).

3.2. Depression and postpartum depression

Depression is a complex disorder that particularly involves neurotransmission processes, especially serotonin receptors and membrane transporters (Meltzer, 1990). The aetiology of the illness is multi factorial, influenced by genetic, environmental, and nutritional factors. Support for a nutritional contribution to the disease derives from studies that report an inverse correlation between the level of omega-3 fatty acids (DHA and/or EPA), as measured in either red blood cell phospholipids or adipose tissue, and symptoms of depression (Freeman *et al.*, 2006). An increased ratio of omega-6/omega-3 is frequently observed in patients suffering from depression (Peet *et al.*, 1998; Haag, 2003). Numerous studies carried out over the last few years are involving omega-3 LC-PUFA supplementation with the reduction of many of the symptoms of different forms of depression, including bipolar disorders, postpartum depression (discussed forward), agoraphobia, and anorexia nervosa (Locke & Stoll, 2001; Logan, 2004). EPA supplementation appears to be more effective than DHA supplementation in reducing the symptoms of depression as concluded from a recent meta-analysis (Lin & Su, 2007). Depression and coronary artery disease often occur in the same individuals who frequently have low plasma levels of EPA and DHA and high levels of AA. Omega-3 LC-PUFA supplementation is effective in the prevention of these pathological disorders (Frasure-Smith *et al.*, 2004; Astog *et al.*, 2008). However, the mechanism(s) by which EPA and/or DHA may reduce depression are still unclear, and more research is needed. As discussed, increasing the nutritional levels of omega-3 fatty acids may modify the activity of integral membrane proteins (receptors, ion channel, molecular pups, etc), and/or counteract the proinflammatory action of AA-derived eicosanoids.

Depression during pregnancy and postpartum depression have negative impacts on the development and health of the newborn. Maternal stress in humans is associated with foetal hypoxia, reduced gestational age, and low birth weight (Wadhwa *et al.*, 1993). Evans *et al* (2001) found that

13.5% of women (n = 14,541) experienced serious symptoms of depression during pregnancy and postpartum. A cross-national analysis of seafood consumption, and the DHA content of breast milk, demonstrated an inverse correlation with the prevalence of pregnancy and postpartum depression. The prevalence varied from 0.5% in Singapore to 24.5% in South Africa, with a mean prevalence rate worldwide of 12.4%. Both higher national seafood consumption and higher DHA content in the mother's breast milk predicted a lower prevalence of postpartum depression. The mean DHA intake of western women is estimated at 15-20 mg/day, whereas intake in countries with high fish consumption (e.g. Japan, Korea, and Norway) is approximately 1000 mg/day. During the third gestational trimester, the foetus accumulates an average of 67 mg/day of DHA, in excess of the dietary intake of many women. Such transfer to the baby through the placenta and, subsequently through breast milk poses a risk to women of significant depletion of omega-3 fatty acids during pregnancy and lactation, thus contributing to the perinatal risk of depression. A review by Parker *et al.*, (2006) about omega-3 fatty acids and postpartum depression, proposed that DHA supplementation in the perinatal period may have additional benefits for the infant's neurodevelopment. Women and their physicians prefer options to standard antidepressant medication during pregnancy and postpartum. DHA supplementation during these periods may be a plausible alternative (Freeman *et al.*, 2006). However, more clinical trials are needed to confirm the recommendation of omega-3 fatty acid supplementation to avoid or reduce symptoms of depression.

3.3. Schizophrenia

Schizophrenia is a psychiatric disease that affects ~1% of the population with a higher prevalence in males than in females. The predominant hypothesis regarding the pathophysiology of the disease is dysfunction of the dopaminergic system. However, further findings concerning the disease suggested a close relationship with reduced tissue levels of omega-6 and omega-3 fatty specially AA and DHA (Yao *et al.*, 2000). Several mechanisms could explain these deficits, including an increased activity of phospholipase A2 thus inducing the extraction of AA and DHA from cerebral membranes (Horrobin *et al.*, 1994). Another argument in favour of a relationship between schizophrenia and omega-6/omega-3 fatty acids is that dietary supplementation of either AA and DHA or their precursors is able to alleviate symptoms of the disease (Arvindakshan *et al.*, 2003). It has been recently proposed that an alteration of DHA metabolism in the brain is involved in the pathophysiology of schizophrenia and that omega-3 fatty acid supplementation may be an important

coadjutant in the treatment of the disease (Kale *et al.*, 2008). It seems therefore that schizophrenia might be an example of a disease in which omega-6 and omega-3 supplementation, presumably AA and DHA, associated with pharmacological treatment might be beneficial, although extended evaluation of such treatment is still required (Fenton *et al.*, 2000).

3.4. Retinal function

Retinal pathologies are not directly involved with mood disorders. However, the retina has neural origin and DHA is essential for the proper development and functioning of this tissue, particularly at the outer membrane segments of photoreceptors cells. DHA is required for the survival of retinal photoreceptors and exerts a protective effect on apoptosis of these photoreceptors during development (Kim *et al.*, 2000). Retinitis pigmentosa is a visual disease with a worldwide prevalence of about 1 in 4000 persons (Boughman *et al.*, 1980). A correlation between retinitis pigmentosa and low retinal DHA levels has been observed, were there is some evidence that the synthesis of DHA is impaired in patients suffering from X-linked retinitis pigmentosa (Kim *et al*, 2000). Supplementation with DHA (400 mg/day) for four years produced a significant a reduction in the loss of the functionality of rods in patients with retinitis pigmentosa, as assessed by an electroretinogram which measures photoreceptor functioning. For patients with retinitis pigmentosa beginning vitamin A therapy, along with DHA (1200 mg/day) slowed the course of the decline in visual field sensitivity (Berson *et al.*, 2004). These results suggest that early intervention with omega-3 fatty acids, presumably with DHA, may be important in slowing down the progression of retinitis pigmentosa.

4. POSSIBLE MECHANISMS FOR LINKS BETWEEN DHA AND MOOD DISORDERS

Several neurophysiological mechanisms have been proposed to explain the relationship between omega-3 polyunsaturated fatty acids and mood disorders (Mamalakis *et al.*, 2002). EPA and DHA appear to decrease the production of inflammatory eicosanoids from AA by means of two mechanisms: First, they compete with AA for incorporation into membrane phospholipids, thus decreasing both cellular and plasma levels of AA. Second, EPA and presumably DHA compete with AA for the cyclo-oxygenase enzyme system, inhibiting the production of proinflammatory eicosanoids derived from AA (e.g. prostaglandins, leukotrienes, hromboxanes). Prostaglandin E2 and thromboxane B2 have been linked to depression. DHA also inhibits the release of proinflammatory cytokines such as interleukin-1 beta, interleukin-2, interleukin-6, interferon gamma, and tumor necrosis factor

alpha which depends on eicosanoid release and are also associated with mood disorders, such as depression (Logan, 2003). Another possible mechanism relates to the abundance of DHA in brain phospholipids were they play a vital role in maintaining the integrity and fluidity of neuronal membranes. By varying the lipid concentration in cell membranes, changes in fluidity can affect either the structure or functioning of proteins embedded in the membrane, including enzymes, receptors or ion channels, leading to changes in cellular signalling (Yehuda *et al.*, 1998; Stahl *et al.*, 2008). Support for the involvement of DHA in receptor functioning, neurotransmitter levels and the metabolism of monoamines implicated in mood disorders has been provided by animal studies (Hibbeln & Salem, 1995).

The hypothesis that DHA can affect membrane fluidity is supported by a recent study using magnetic resonance imaging (Hirashima *et al.*, 2004). A group of bipolar women received DHA for 4 weeks and were contrasted with a non treated bipolar group. T_2 whole relaxation times were used to detect changes in membrane fluidity, measured at baseline and 4 weeks after omega-3 fatty acid treatment. The bipolar patients receiving DHA treatment showed significantly decreased T_2 values, with a dose-dependent effect compared to the control group. Another hypothesis involves a more direct mechanism controlling gene expression and the binding of fatty acids to specific nuclear receptors early in life, as was discussed above, leading to genetic transcription and predisposure to a range of diseases related to DHA depletion later in life, such as Alzheimer's disease, cardiac disease, and depression (Sampath & Ntambi, 2004).

5. CONCLUSIONS AND FUTURE PROSPECTS

A significantly amount of references clearly establish that DHA is important for the proper neurodevelopment of the brain and visual system. Epidemiological evidence suggest that a decrease in brain DHA levels, which normally occurs during aging, and that is exacerbated by reduced dietary intake of DHA, may increase the prevalence of several neurological diseases, such as Alzheimer's disease. However, at present we do not understood at all the complex functions that DHA performs as either a free fatty acid or incorporated into phospholipids. Further research is needed to better understand the process of DHA transport, internalization and membrane inclusion, as well as DHA metabolism into its incorporation into phospholipids. The identification of several DHA-derived metabolites, probably involved in cell signalling suggests that free DHA is utilized to perform many functions beyond a structural role in phospholipids and membrane structure. Future research about food and/or additives that preferentially provide DHA and molecules that promote its internalization, transport and metabolism is clearly needed to fully understand the importance of DHA in the development, normal function, senescence, and pathology of the nervous system. Establishing the functions of DHA in the nervous system will be critical to appreciating the possible health implications of a reduced dietary intake of DHA currently occurring in western populations, and the importance of DHA supplementation, at the early and late stages of human life. Basic, clinical and epidemiologic research supports a protective effect of omega-3 LC-PUFA, particularly of DHA in mood disorders. However, results are highly heterogeneous and in some cases confounding, indicating that it is important to examine the results of each individual study in order to anticipate more general conclusions about the role of DHA in the brain and nervous system and in mood disorders.

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