

n-3 Polyunsaturated Fatty Acids Decrease Anxiety Feelings in a Population of Substance Abusers

Laure Buydens-Branchey, MD and Marc Branchey, MD

Abstract: There is mounting evidence that low levels of n-3 polyunsaturated fatty acids (PUFAs) play a role in the pathophysiology of a number of psychiatric disorders. Preclinical studies have shown that n-3 PUFAs decrease anxietylike behaviors, but there is a paucity of information about their effects on anxiety in humans. In light of our observation that substance abusers have poor dietary habits and the strong association between anxiety disorders and substance use disorders, the possibility that the administration of supplements of n-3 PUFAs would decrease the anxiety level of a group of substance abusers was explored.

Thirteen patients were given on a daily basis capsules containing 3 g of n-3 PUFAs (eicosapentaenoic acid + docosahexaenoic acid). Eleven patients received similarly looking placebo capsules containing vegetable oil. The trial was double-blind, randomized, and lasted 3 months. A scale assessing anxiety feelings was administered at baseline and on a monthly basis thereafter. Six PUFA group patients and 8 placebo group patients were followed for an additional 3 months after treatment discontinuation and administered the same questionnaire monthly.

Patients who received n-3 PUFAs for 3 months showed a progressive decline in anxiety scores. This was not the case for patients who received placebos. A comparison of the 2 groups was significant ($P = 0.010$). Anxiety scores remained significantly decreased in the PUFA group for 3 months after treatment discontinuation. A comparison of the 2 groups followed for 6 months was also significant ($P = 0.042$).

In conclusion, these preliminary data indicate that n-3 PUFA supplementation could be beneficial in the treatment of some patients with anxiety disorders.

(*J Clin Psychopharmacol* 2006;26:661–665)

Preclinical studies have indicated that the administration of n-3 polyunsaturated fatty acids (PUFAs) decreases anxietylike behaviors in rodents. Interleukin 1β , the most potent proinflammatory cytokine, was found to induce an inflammatory sickness response, stress and anxietylike behaviors, and a stimulation of corticosterone secretion in rats. These changes were significantly reversed by the

administration of the long-chain n-3 PUFA, eicosapentaenoic acid (EPA, 20:5n-3).^{1,2}

Although human studies have shown that n-3 supplementation was beneficial in a variety of psychiatric disorders, including depression, aggression, attention-deficit/hyperactivity disorder (ADHD), borderline personality disorder, and dementia,^{3,4} there is a limited amount of information about their effects in anxiety disorders. Yehuda et al who investigated the effects of the administration of a mixture of n-3 and n-6 PUFAs on test anxiety in college students observed an improvement in variables associated with this type of anxiety (ie, appetite, mood, concentration, fatigue, academic organization, and sleep). They also observed a decrease in elevated cortisol level.⁵ Fux et al,⁶ on the other hand found EPA to be ineffective in a preliminary study of 11 patients with obsessive-compulsive disorder treated with selective serotonin reuptake inhibitors.

In light of the reported effectiveness of EPA in decreasing anxietylike behaviors in preclinical studies and of the scarcity of human data, a double-blind, placebo-controlled trial of n-3 PUFAs on anxiety symptoms in humans seemed warranted. It could be hypothesized that individuals whose n-3 PUFAs intake is deficient would more readily respond to n-3 supplementation than those whose intake is closer to recommended amounts. Depressive disorders and homicides, believed to be associated with n-3 PUFA deficiencies, are high in countries where the consumption of foods rich in these nutrients, such as fish, is low.^{7,8} We had observed that the diets of substance abusers are less than optimal. Moreover, the study of patterns of comorbidity between psychiatric conditions indicates that there is a strong association between anxiety disorders and substance use disorders.⁹ For these reasons, the subjects selected as participants in this study were substance abusers. They were treated for 3 months with a combination of EPA and another long-chain n-3 PUFA, docosahexaenoic acid (DHA, 22:6n-3), because the bulk of the literature so far points to the efficacy of such a combination in improving psychiatric symptoms, whereas DHA alone appears to be less effective. It was hypothesized that patients' consumption of n-3 PUFA-containing foods before the start of the study would be below recommended amounts and that the administration of long-chain n-3 PUFA supplements would decrease their anxiety level.

MATERIALS AND METHODS

Patients

Twenty-four men attending the New York Harbor Healthcare System (Brooklyn site) substance abuse clinics

New York Harbor Healthcare System, Brooklyn Campus, Brooklyn, NY. Received May 8, 2006; accepted after revision September 18, 2006. This work was supported by grant R01-DA15360 from the National Institute on Drug Abuse.

Address correspondence and reprint requests to Laure Buydens-Branchey, MD, New York Harbor Healthcare System, Brooklyn Campus, 151/BK, 800 Poly Place, Brooklyn, NY 11209. E-mail: lbuydens@worldnet.att.net.

Copyright © 2006 by Lippincott Williams & Wilkins

ISSN: 0271-0749/06/2606-0661

DOI: 10.1097/01.jcp.0000246214.49271.fl

were selected for participation in this study. They were included if it was determined that they did not present any major physical illness on the basis of a physical examination and after the administration of a battery of laboratory tests. Patients were interviewed by a psychiatrist and administered the Addiction Severity Index questionnaire.¹⁰ The Addiction Severity Index explores the following areas: alcohol and drug use, psychiatric status, family history of alcohol, drug, and psychiatric problems, family and social relationships, medical status, legal status, and employment/support status. Patients were included if they did not present or did not have a history of schizophrenia, major depressive disorder, bipolar disorder, mental retardation, or dementia. If subjects were judged to be appropriate for enrollment, written informed consent was obtained after a description of the study. The trial was double blind, and assignment to the active substance group or to the placebo group was random.

Initial Assessment

The following assessments covering the year and the month preceding the study entry were obtained: nature of the substances used, amount of each substance used, and frequency of use. A diet questionnaire covering the month preceding the study entry was obtained. This questionnaire was an adaptation of the National Institutes of Health Diet History Questionnaire.¹¹ Based on the results of this questionnaire, each patient's energy (kilocalories) and n-6 and n-3 PUFA intake were calculated, using the US Department of Agriculture National Nutrient Database Reference.¹² A modified version of the Profiles of Mood States (POMS)¹³ was administered. The POMS is a self-report questionnaire that requires subjects to rate the intensity of 65 mood items on a 5-point scale. Ratings are combined into 6 scores assessing vigor, depression, tension, anger, confusion, and fatigue. It was modified for use in this study because we had observed that when patients attending our clinics were asked to make 325 choices when given the complete POMS, they quickly lost interest in the task and gave unreliable answers. The modified POMS version was limited to the 36 items comprising the anger, tension, and depression subscales, with the following 2 choices for each item: present or absent. This reduced the total number of choices to 72, making the questionnaire more readily acceptable. Changes in tension scores only are the topic of the present report.

Polyunsaturated Fatty Acid and Placebo Treatment

Patients were treated for 3 months. They received either capsules containing n-3 PUFAs or placebo capsules. Both types of capsules were purchased from Nordic Naturals (Watsonville, Calif). The PUFA capsules contained 450 mg of EPA, 100 mg of DHA, and 50 mg of other n-3 PUFAs (triglycerides). The placebo capsules contained vegetable oil (soybean). Both types of capsules contained small amounts of lemon oil to mask their taste. Patients were advised to take 5 capsules each day. Those taking the active substances were thus given a daily amount of 3 g of long-chain n-3 PUFAs.

Follow-up Assessments

Thirteen patients assigned to the PUFA group and 11 patients assigned to the placebo group were available for follow-up during the 3-month treatment period. One, 2, and 3 months after the start of the study, dietary questionnaire and modified POMS covering the 1-month period that elapsed between the 2 interviews were administered to all patients.

A subset of 6 patients in the PUFA group and 8 patients in the placebo group was followed for an additional period of 3 months after treatment had been discontinued and administered the questionnaires on a monthly basis. This last group of 14 patients was thus followed for a total of 6 months.

To ensure compliance, patients were asked to bring back their empty capsule containers at the time of their monthly interviews before receiving a new batch of capsules. They were asked whether they had experienced any side effect after the ingestion of the capsules.

Statistical Analysis

Comparisons of the 2 patient groups' demographic and baseline dietary data were made with 2-tailed Student *t* tests, χ^2 tests with Yates continuity correction or Fisher exact tests, as appropriate.

Comparisons of the 2 patient groups' tension scale baseline values were done with Student *t* tests.

Comparisons of the changes over time in the tension scale of the patient groups were done with repeated measures analyses of covariance (ANCOVAs) with baseline values as covariates. Because of the small number of study participants and the resulting low statistical power of the analyses, effect sizes (*f*) for the differences between the 2 groups were also calculated. Small, medium, and large effect sizes correspond to *f* values of 0.10, 0.25, and 0.50, respectively.¹⁴

RESULTS

Patient Characteristics

The PUFA and placebo groups did not differ significantly in age (52.0 ± 7.0 vs 50.3 ± 7.0 years). There were no significant differences either in marital status, educational level, employment status, or types of drugs used for a period of 1 year before the start of the study. Drugs used were alcohol, cocaine, and heroin, taken either alone or in various combinations. Nine patients (5 in the PUFA group and 4 in the placebo group) were maintained on doses of methadone ranging from 70 to 120 mg. These doses remained stable throughout the study. Five patients in each group were treated with antidepressants. Doses of antidepressants remained stable throughout the study.

Side Effects

None of the study participants reported side effects except for 3 patients who stated having loose stools.

Dietary Data

Dietary data are shown in Table 1. The mean daily fish intake of the 24 study participants was 35.7 ± 18.6 g. Their mean daily n-6 PUFA intake was 16.0 ± 8.8 mg, and their

TABLE 1. Baseline Daily Calories, Fish, and PUFA Intakes

	All Patients (n = 24)	n-3 PUFAs (n = 13)	Placebo (n = 11)	P
Energy (kcal)	2286 ± 1424	2305 ± 1499	2261 ± 1401	NS
Fish (g)	35.7 ± 18.6	35.9 ± 20.9	35.5 ± 16.4	NS
Total PUFAs (g)	17.41 ± 9.62	18.39 ± 10.06	16.14 ± 9.38	NS
n-6 PUFAs (g)				
Total	15.96 ± 8.84	16.91 ± 9.18	14.73 ± 8.70	NS
Short chain (C18)	15.72 ± 8.71	16.70 ± 9.04	14.45 ± 8.56	NS
Long chain (C20, C22)	0.24 ± 0.23	0.21 ± 0.19	0.28 ± 0.28	NS
n-3 PUFAs (g)				
Total	1.45 ± 0.86	1.48 ± 0.96	1.40 ± 0.78	NS
Short chain (C18)	1.30 ± 0.78	1.35 ± 0.86	1.24 ± 0.71	NS
Long chain (C20, C22)	0.15 ± 0.10	0.13 ± 0.11	0.16 ± 0.08	NS
Daily intake of long-chain n-3 PUFAs as percentage of ISSFAL recommended intake	30 ± 19	27 ± 22	33 ± 16	NS

Values are means ± SD. Comparisons between the PUFA and placebo groups were made with 2-tailed Student *t* tests.

mean daily n-3 PUFA intake was 1.45 ± 0.86 mg. Patients consumed thus 11 times more n-6 than n-3 PUFAs. There were no significant differences between the PUFA and control groups in mean daily energy (kilocalories), mean daily amounts of fish, mean daily amounts of total, short-chain, or long-chain n-6 PUFAs, and total, short-chain, or long-chain n-3 PUFAs consumed during the month preceding the start of the study. The fish most frequently consumed were catfish, whiting, flounder, or canned tuna. Calculations of differences between the daily intake of 500 mg of long-chain n-3 PUFAs, recommended by the International Society of the Study of Fatty Acids and Lipids (ISSFAL),¹⁵ and patients' actual daily intake did not reveal differences between the groups but showed that both groups fell short of the recommended intake. The PUFA group patients consumed on average 27% and the placebo group 33% of this recommended intake.

Effects of n-3 PUFAs and Placebo on Tension Scale Scores

Figure 1 illustrates baseline and changes in the tension scale scores during the 3-month administration of n-3 PUFAs or placebo capsules. The difference between baseline values assessed by *t* test was not significant. The figure shows a decline in tension scores in the PUFA group but not in the placebo group. A comparison of the 2 groups by repeated measures ANCOVA (with baseline values as covariates) revealed a significant difference ($F_{1,21} = 8.070$, $P = 0.010$) and a large effect size ($f = 0.62$).

Figure 2 illustrates baseline and changes in the tension scale scores of the patients who were followed for 6 months. The difference between baseline values assessed by *t* test was not significant. Scores remained decreased in patients assigned to the PUFA group from month 4 through month 6 after treatment discontinuation. A comparison of the 2 patient groups who were followed for 6 months by repeated measures ANCOVA (with baseline values as covariates)

revealed a significant difference ($F_{1,11} = 5.269$, $P = 0.042$) and a large effect size ($f = 0.69$).

DISCUSSION

In this study, the daily administration of 3 g of long-chain n-3 PUFAs (EPA + DHA) for a period of 3 months significantly decreased scores on a tension scale in a group of substance abusers. These scores did not return to pretreatment levels after treatment discontinuation. Reasons for the persistence of the antianxiety effects after discontinuation of n-3 supplementation are not known. One possibility

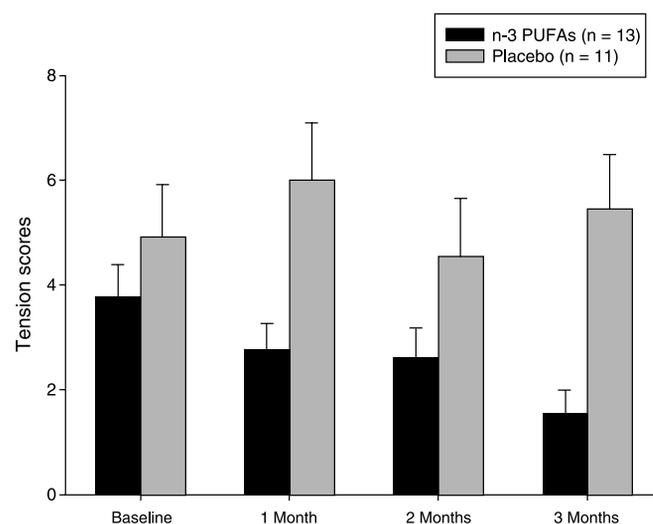


FIGURE 1. Tension scale scores (mean and SEM) of substance abusers during a 3-month administration of n-3 PUFAs or placebo. A comparison of the scores of the 2 patient groups by a repeated measures ANCOVA (with baseline values as covariates) revealed a significant difference ($P = 0.010$). The effect size was large ($f = 0.62$).

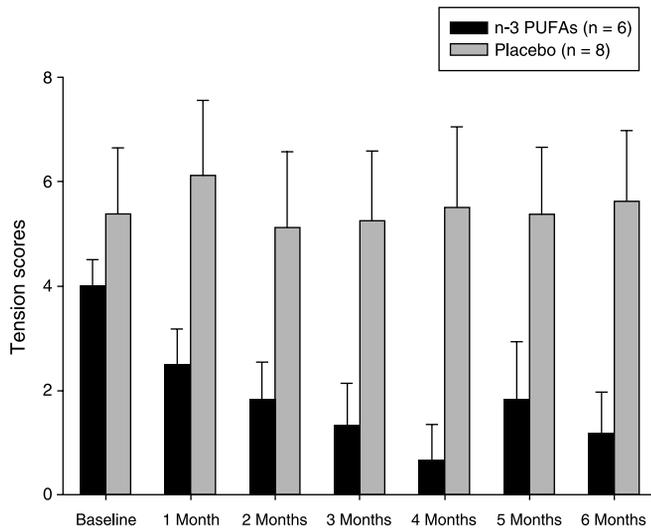


FIGURE 2. Tension scale scores (mean and SEM) of substance abusers during a 3-month administration of n-3 PUFAs or placebo and a follow-up period of 3 months (months 4–6), during which no capsules were administered. A comparison of the scores of the 2 patient groups by a repeated measures ANCOVA (with baseline values as covariates) revealed a significant difference ($P = 0.042$). The effect size was large ($f = 0.69$).

is that, in the course of the present study, patients' intake of long-chain n-3 PUFAs that were approximately 20 times larger than those provided by their usual diet induced CNS changes that were not reversed immediately after the interruption of treatment. It is also possible that some of the fatty acids surplus was stored in fatty tissues and continued to leach out of these stores for a few months.

On the basis of epidemiological and cross-sectional studies, there is little argument that n-3 deficiencies are associated with astonishingly diverse psychiatric disorders that include, but are not limited to, depression, aggression, ADHD, borderline personality disorder, schizophrenia, autism, dementia, and substance abuse.^{3,4,16,17} Interventional studies on the whole support conclusions derived from epidemiological and cross-sectional studies. Supplements of EPA, a combination of EPA and DHA, and, in some studies, DHA alone were found to improve the psychiatric symptoms of patients with mood disorders, ADHD, and schizophrenia.^{3,4} The present results should be considered as preliminary. If confirmed by other studies, anxiety disorders could be added to the list of disorders affected by n-3 PUFAs.

The apparent multitude of PUFA effects could be understood in light of their extraordinary diversity of actions. They are divided into 2 major families, the n-3 and the n-6 families. EPA and DHA are the main long-chain derivatives of the precursor ALA (α -linolenic acid, 18:3n-3). Arachidonic acid (AA, 20:4n-6) is the main long-chain derivative of the n-6 series precursor linoleic acid (18:2 n-6). Some PUFAs, mostly AA and DHA, and, to a lesser extent, EPA are components of neuronal membranes phospholipids and modulate membranes dynamic properties. Changes in

the composition of these phospholipids can affect neurotransmitter release, neurotransmitter receptor function, ion channel function, and enzyme activity. Upon stimulation, both n-6 and n-3 PUFAs are cleaved from membrane phospholipids under the influence of phospholipases and are converted via different pathways to mediators that have opposing effects. Arachidonic acid-derived mediators are vasoconstrictive, proaggregant, and proinflammatory, whereas EPA-derived mediators have vasodilating, antiaggregating, and anti-inflammatory actions. Finally, PUFAs and their metabolites regulate gene transcriptions. One could wonder why EPA, which is not abundant in neuronal membranes, appears more effective than DHA alone. It has been suggested that EPA could play a role in brain function by counteracting the AA-mediated signaling. It could, for example, competitively attenuate the formation of the n-6-derived eicosanoids that mediate immune-inflammatory responses that have been linked to the pathophysiology of depressive disorders, bipolar disorders, and schizophrenia.^{18,19} EPA could also counteract the vasoconstrictive effects of AA,³ thereby increasing blood flow in the brain.

Recommendations for the intake of n-3 PUFAs necessary to cover human requirements are still in the process of being determined and will have to await the completion of additional trials. These recommendations vary from country to country and depend in part on the amounts of n-6 PUFAs present in different diets. Based on a review of major epidemiological studies conducted in the United States, the daily intake of EPA and DHA recommended by ISSFAL¹⁵ for cardiovascular health is 500 mg. Participants in the present study consumed only, on average, 149 mg of long-chain n-3 PUFAs daily or 30% of the ISSFAL recommendation. Their intake of EPA and DHA was most likely even lower because in our calculations of long-chain n-3 PUFAs, docosapentaenoic acid (22:5n-3) was included. This intake, albeit low, lies however within the range of mean long-chain n-3 PUFAs intake found in other population samples in this country,^{20,21} indicating that large segments of the US population probably do not consume adequate amounts of n-3 PUFAs.

Our interpretation of the data should be taken in the context of our study's possible limitations. The number of participants was small, and some of them received medications. It should be pointed out that medication doses remained stable during the study and that a number of other studies of the efficacy of n-3 PUFAs supplementation in mood disorders involved individuals on standard medication who experienced further improvements that were attributed to the PUFA supplements.

In conclusion, in this preliminary study, the daily administration of 3 g of long-chain n-3 PUFAs for a period of 3 months significantly decreased anxiety feelings in comparison with the administration of a placebo. This decrease persisted for 3 months after treatment discontinuation. Data on the efficacy of n-3 PUFAs in anxiety disorders are almost nonexistent. The present data need to be replicated and could lead to the use of treatments or treatment adjuncts in some individuals presenting anxiety symptoms and whose n-3 PUFA intakes are deficient.

REFERENCES

1. Song C, Li X, Leonard BE, et al. Effects of dietary n-3 or n-6 fatty acids on interleukin-1beta-induced anxiety, stress, and inflammatory response in rats. *J Lipid Res.* 2003;44:1984–1991.
2. Song C, Leonard BE, Horrobin DF. Dietary ethyl-eicosapentaenoic acid but not soybean oil reverses central interleukin-1-induced changes in behavior, corticosterone and immune response in rats. *Stress.* 2004;7:43–54.
3. Alessandri JM, Guesnet P, Vancassel S, et al. Polyunsaturated fatty acids in the central nervous system: evolution of concepts and nutritional implications throughout life. *Reprod Nutr Dev.* 2004;44:509–538.
4. Young GS, Conquer J. Omega-3 fatty acids and neuropsychiatric disorders. *Reprod Nutr Dev.* 2005;45:1–28.
5. Yehuda S, Rabinovitz S, Mostofsky DI. Mixture of essential fatty acids lowers test anxiety. *Nutr Neurosci.* 2005;8:265–267.
6. Fux M, Benjamin J, Nemets B. A placebo-controlled cross-over trial of adjunctive EPA in OCD. *J Psychiatr Res.* 2004;38:323–325.
7. Hibbeln JR. Fish consumption and major depression. *Lancet.* 1998; 351:1213.
8. Hibbeln JR, Salem N Jr. Omega-3 fatty acids and psychiatric disorders. Current status of the field. In: Mostofsky D, Yehuda S, Salem N Jr, eds. *Fatty Acids: Physiological and Behavioral Functions.* Totowa, NJ: Humana Press; 2001:311–330.
9. Merikangas KR, Metha RL, Molnar BE, et al. Comorbidity of substance use disorder with mood and anxiety disorders: results of the International Consortium in Psychiatric Epidemiology. *Addict Behav.* 1998; 23:893–907.
10. McLellan AT, Kushner H, Metzger D, et al. The fifth edition of the Addiction Severity Index. *J Subst Abuse Treat.* 1992;9:199–213.
11. National Cancer Institute, US National Institutes of Health. *Diet History Questionnaire.* Available at: <http://riskfactor.cancer.gov/DHQ/>. Accessed 2005.
12. US Department of Agriculture, Agricultural Research Service. *USDA National Nutrient Database for Standard Reference, Release 18.* [Nutrient Data Laboratory Home Page]. Available at: <http://www.nal.usda.gov/fnic/foodcomp>. Accessed 2005.
13. McNair DM, Lorr M, Droppleman LF. *Manual for the Profile of Mood States.* San Diego, CA: Educational Testing Service; 1971.
14. Cohen J. *Statistical Power Analysis for the Behavioral Sciences.* 2nd ed. Hillsdale, NJ: Lawrence Erlbaum; 1988.
15. International Society for the Study of Fatty Acids and Lipids. *Recommendations for Dietary Intake of Polyunsaturated Fatty Acids in Healthy Adults.* Brighton, UK: International Society for the Study of Fatty Acids and Lipids; 2004.
16. Buydens-Branchey L, Branchey M, McMakin DL, et al. Polyunsaturated fatty acid status and aggression in cocaine addicts. *Drug Alcohol Depend.* 2003;71:319–323.
17. Buydens-Branchey L, Branchey M, McMakin DL, et al. Polyunsaturated fatty acid status and relapse vulnerability in cocaine addicts. *Psychiatry Res.* 2003;120:29–35.
18. Maes M, Stevens WJ, Declerck LS, et al. Significantly increased expression of T-cell activation markers (interleukin-2 and HLA-DR) in depression: further evidence for an inflammatory process during that illness. *Prog Neuropsychopharmacol Biol Psychiatry.* 1993;17:241–255.
19. Maes M, Bosmans E, Calabrese J, et al. Interleukin-2 and interleukin-6 in schizophrenia and mania: effects of neuroleptics and mood stabilizers. *J Psychiatr Res.* 1995;29:141–152.
20. Ma J, Folsom AR, Shshar E, et al. Plasma fatty acid composition as an indicator of habitual dietary fat intake in middle-aged adults. *Am J Clin Nutr.* 1995;62:564–571.
21. Iribaren C, Markovitz JH, Jabobs DR Jr, et al. Dietary intake of n-3, n-6 fatty acids and fish: relationship with hostility in young adults—the CARDIA study. *Eur J Clin Nutr.* 2004;58:24–31.