Effects of n–3 long-chain polyunsaturated fatty acids on depressed mood: systematic review of published trials[1–3]

Katherine M Appleton, Robert C Hayward, David Gunnell, Tim J Peters, Peter J Rogers, David Kessler, and Andrew R Ness

ABSTRACT

Background: Greater dietary intakes of n−3 long-chain polyunsaturated fatty acids (n–3 PUFAs) may be beneficial for depressed mood.

Objective: This study aimed to systematically review all published randomized controlled trials investigating the effects of n–3 PUFAs on depressed mood.

Design: Eight medical and health databases were searched over all years of records until June 2006 for trials that exposed participants to n–3 PUFAs or fish, measured depressed mood, were conducted on human participants, and included a comparison group.

Results: Eighteen randomized controlled trials were identified; 12 were included in a meta-analysis. The pooled standardized difference in mean outcome (fixed-effects model) was 0.13 SDs (95% CI: 0.01, 0.25) in those receiving n–3 PUFAs compared with placebo, with strong evidence of heterogeneity ($I^2 = 79\%$, $P < 0.001$). The presence of funnel plot asymmetry suggested that publication bias was the likely source of heterogeneity. Sensitivity analyses that excluded one large trial increased the effect size estimates but did not reduce heterogeneity. Metaregression provided some evidence that the effect was stronger in trials involving populations with major depression—the difference in the effect size estimates was 0.73 (95% CI: 0.05, 1.41; $P = 0.04$), but there was still considerable heterogeneity when trials that involved populations with major depression were pooled separately ($I^2 = 72\%$, $P < 0.001$).

Conclusions: Trial evidence that examines the effects of n–3 PUFAs on depressed mood is limited and is difficult to summarize and evaluate because of considerable heterogeneity. The evidence available provides little support for the use of n–3 PUFAs to improve depressed mood. Larger trials with adequate power to detect clinically important benefits are required. *Am J Clin Nutr* 2006;84:1308–16.

KEY WORDS n−3 Long-chain polyunsaturated fatty acids, depressed mood, randomized controlled trials, systematic review

INTRODUCTION

n−3 Long-chain polyunsaturated fatty acids (n–3 PUFAs) are essential fatty acids implicated in the development of several human conditions and diseases, including depression (1). Several lines of evidence suggest there is a relation in humans between dietary intake of n–3 PUFAs and depressed mood.

There is biochemical evidence that n–3 PUFAs play an important role in neural structure and function. The brain and central nervous system contain high concentrations of n–3 PUFAs, and several studies suggest a role for n–3 PUFAs in neurotransmitter synthesis, degradation, release, reuptake, and binding (2–4). Low concentrations of n–3 PUFAs have been associated with lower concentrations of the neurotransmitter dopamine, lower density of neurotransmitter receptors D_2, lower binding of D_2 receptors (2–4), and increased serotonin activity and increased density of 5-HT_{2A} receptors in the frontal cortex (2–4).

Epidemiologic evidence is also available from ecological and cross-sectional studies. Hibbeln (5) showed a strong negative association between fish intake and depression across 13 countries. Tanskanen et al (6) reported a higher prevalence of depressive symptoms in infrequent than in frequent fish consumers in Finland, Silvers and Scott (7) found lower mental health status in non-fish consumers than in fish consumers in New Zealand, and Timonen et al (8) showed an increased risk of developing depression in persons who rarely ate fish compared with regular fish eaters, although effects were only found for females.

A relation between n–3 PUFA intake and depressed mood has also been reported in clinical studies. Lower concentrations of n–3 PUFAs have been reported in the plasma or red blood cell membranes of persons with a Diagnostic and Statistical Manual of Mental Disorders 4th edition major depressive disorder diagnosis compared with matched nondepressed control subjects (9, 10). Depression severity has also been found to correlate positively with balance between n–3 PUFA and n–6 long-chain PUFAs in plasma and erythrocyte phospholipids (11, 12).

More recently, several trials have reported a beneficial effect of n–3 PUFA supplementation on depression in clinical populations. For example, Stoll et al (13) observed improvements in the depressive symptoms associated with bipolar disorder after supplementation with n–3 PUFAs compared with placebo, Nemets et al (14) reported benefits of n–3 PUFAs compared with placebo for treating unipolar depressive disorder, and Peet and Horrobin (15) found n–3 PUFAs to be effective in treating

[1] From the Department of Experimental Psychology (KMA, RCH, and PJR), the Department of Social Medicine (DG and ARN), and the Academic Unit of Primary Health Care, Department of Community Based Medicine (TJP and DK), University of Bristol, Bristol, United Kingdom.

[2] Supported by the Food Standards Agency, Government of the United Kingdom (grant no. NO5038) and The University of Bristol, United Kingdom.

[3] Reprints not available. Address correspondence to KM Appleton, School of Psychology, Queen's University, Belfast, 18-30 Malone Road, Belfast, BT9 5BP United Kingdom. E-mail: k.appleton@qub.ac.uk.

Received February 9, 2006.

Accepted for publication August 14, 2006.

ongoing depression that remained unresponsive to standard therapies. These trials, however, have generally been small. Furthermore, not all studies have reported positive effects. Fenton et al (16) found no benefits of n–3 PUFAs on depressed mood compared with placebo for patients with schizophrenia, and Llorente et al (17) found no clinical improvement in patients with postpartum depression using n–3 PUFAs compared with placebo. The aim of the present review was to identify and combine all published randomized controlled trials investigating the effects of dietary supplementation with n–3 PUFAs on depressed mood.

METHODS

Published randomized controlled trials were identified by searching databases. Trials were obtained, and relevant data were abstracted, tabulated, and formally combined.

Identification of potentially relevant reports

Eight databases were searched over all years of records until June 2006. These databases were the following: MEDLINE, EMBASE, PsycInfo, CINAHL, Biosis, AMED, the Cochrane Controlled Trials Register, and the Cochrane Database of Systematic Reviews. Articles investigating the effects of n–3 PUFAs on depressed mood were identified by using the following search terms for n–3 PUFAs: "n−3," "omega-3," "ω-3," "essential fatty acid," "ALA," "α-linolenic acid," "fish," "fatty fish," and "cod liver oil." These were combined with the following search terms for depressed mood: "depression," "depressive disorder," "depressed mood," "mood," "mood disorder," "affective disorder," "affect," "anxiety," "postpartum," "involutional," "dysthymic disorder," "seasonal affective disorder," in either the "Keyword" or "Abstract" sections of all databases. Duplicates were removed. Titles and abstracts were inspected independently by 2 researchers (KMA and RCH, HLS, or PLB), and articles clearly identified as not relevant were removed from the list. The reference lists of the relevant reports were also inspected to identify any additional trials not identified by the searches.

Report inclusion and data abstraction

Full copies of all relevant articles were acquired where possible over the duration of the data collection period. Some articles were not available through the British Library, so these articles were not acquired. Each article was independently assessed for inclusion in the review by 2 researchers (KMA and RCH, HLS, or PLB), using 5 inclusion criteria. These criteria were the following: exposure was n–3 PUFAs or fish, outcome measures included depressed mood, study was conducted on human participants, study included a comparison group, and reported a randomized controlled trial or a clinical controlled trial. Data were abstracted independently from each identified trial by 3 researchers (KMA; RCH, HLS, or PLB; and DG, DK, TJP, PJR, or ARN) using a standard data abstraction form. Discordances were discussed and resolved. Where data were incomplete, corresponding authors were contacted directly for relevant information.

Study quality

Study quality was assessed by using reports of adequate concealment of treatment allocation, blinding of study participants and researchers, and use of an intention-to-treat analysis.

Data analysis

The methods and results of all trials investigating the effects of n–3 PUFAs on depressed mood were tabulated. All trials reported continuous data (self-report ratings of depression) as opposed to dichotomous data, but trials used many psychometric instruments. Data from trials reporting means (±SDs) were combined (18). To include data from as many trials as possible, missing SD data for one trial were imputed from SD data from all other trials that used the same measure for depression (19), and the standardized mean effect for all trials was calculated by using Hedges' adjusted *g* (20). Hedges' adjusted *g* is a formulation of effect size used in the standardized mean difference method that includes an adjustment to correct for small sample bias (20). Both random- and fixed-effects models were used to estimate the overall effect size. Random-effects models are theoretically preferable when combining the results of studies when heterogeneity exists. However, where the heterogeneity is due to publication bias, fixed-effects models may be preferable because they give less weight to smaller studies (18, 20, 21). Heterogeneity was investigated by using Higgins' I^2 statistic (22, 23). The I^2 statistic describes the proportion of total variation in study estimates that is due to heterogeneity. Three possible sources of heterogeneity were identified a priori—publication bias, the inclusion of one large trial in which depression was not a primary outcome measure, and the inclusion of trials involving different clinical populations. Possible publication bias was investigated by drawing a funnel plot to look for funnel plot asymmetry (24). Heterogeneity as a result of inclusion of one large trial in which depression was not a primary outcome measure was investigated by using sensitivity analyses (20). The effects of trial population were investigated with the use of metaregression (predictor: major or other depressive illness) and sensitivity analyses were also conducted. Analyses were performed in STATA version 8 (StataCorp, College Station, TX) by using the "METAN," "FUNNEL," and "METAREG" commands.

RESULTS

The process of identification and inclusion of trials is summarized in **Figure 1**. The methods of the 18 relevant randomized controlled trials are displayed in **Table 1**. The number of persons enrolled in the trials ranged from 11 (32) to 452 (29). Some trials recruited males only (29), some females only (17, 31, 33), some trials recruited both sexes (13–16, 25–28, 30, 32, 34–37), and all trials involved adults, excepting one conducted on children (37). Trial participants either had a diagnosis of various different clinical conditions, including unipolar depression (14, 15, 28, 30, 34, 37), bipolar disorder (13, 27, 33, 36), schizophrenia (16), chronic fatigue symdrome (25, 26), postpartum depression (17), borderline personality disorder (31), obsessive compulsive disorder (32), and angina (29), or were healthy volunteers (35). The intervention varied considerably between studies. The daily dose ranged from 0.2 g PUFA (17) to 9.6 g PUFA (13) and was composed of eicosapentaenoic acid only (14–16, 27, 31, 32, 36), docosahexanoic acid only (17, 28), or a combination of eicosapentaenoic acid and docosahexanoic acid (13, 25, 26, 29, 30, 33–35, 37). Duration of supplementation ranged from 28 d (14, 33) to 180 d (29). In some studies, n–3 PUFA supplementation was given alone (eg, 26), and in others n–3 PUFA supplementation was given in addition to a range of existing treatments (eg, 31). Depressed mood was also measured differently in

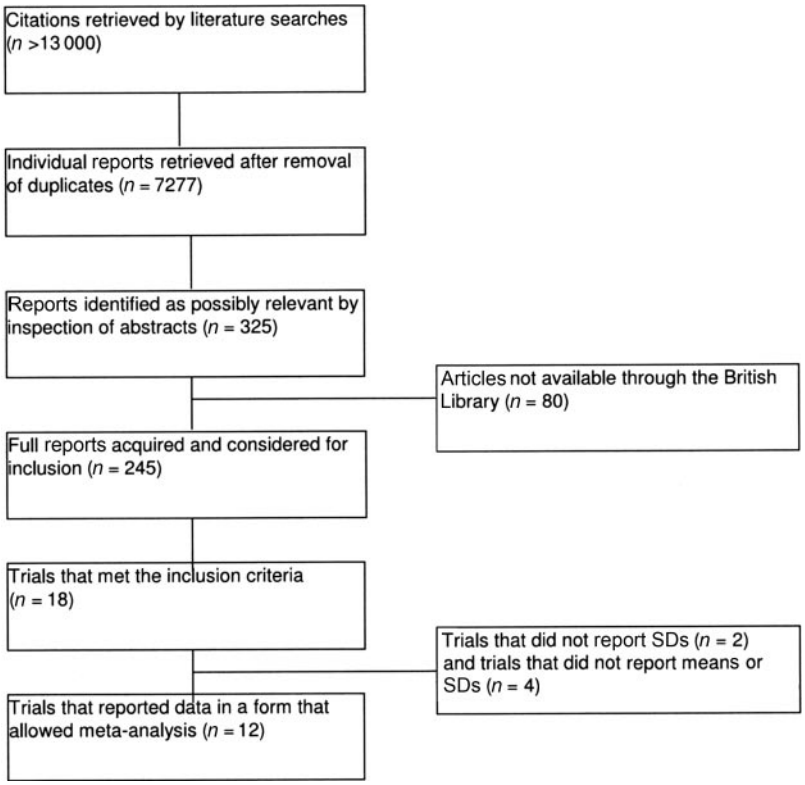

FIGURE 1. Process of inclusion of studies for review and analysis of studies included in the systematic review of n–3 long-chain polyunsaturated fatty acids and depressed mood.

different studies. Most studies used the Hamilton Depression Rating Scale (HDRS; 38)(13–15, 28, 30, 32, 33, 36), the Montgomery-Asberg Depression Rating Scale (MADRS; 39) (15, 16, 28, 31), or the Beck Depression Inventory (BDI; 40) (15, 17, 26, 34), but the short form of the Hamilton Depression Rating Scale (HRDS-SF; 41) (34), the Inventory of Depressive Symptomatology (IDS-C; 42) (27), the Children's Depression Rating Scale (CDRS; 43), the Children's Depression Inventory (CDI; 44), the Clinical Global Impression (CGI; 45) (37), the Depression scale of the Derogatis Stress Profile (DSP; 46) (29), the Depression scale of the Profile of Mood States (POMS; 47) (35), and a 4-point Likert scale (25) were also used. Most studies reported that they had used blinding (13–17, 25–27, 30, 31, 33–37), 8 studies described adequate allocation concealment (14, 16, 17, 26, 34–37), and 9 had clearly used intention-to-treat analyses (15, 17, 28, 29, 32–36). The results, as reported for all studies, are shown in **Table 2**, along with the effect sizes and CIs used in the meta-analyses.

Twelve trials were included in the meta-analyses. Four trials (26, 27, 32, 33) could not be included because no means or SDs were available, and 2 trials (25, 35) could not be included because no SDs were available and could not be imputed because no other studies had used the same measure of depression. SDs were imputed for one trial only (15). Three studies (15, 28, 34) reported results for >1 outcome measure of depressed mood. Results for the HDRS (15, 28) and BDI (34) were used in the meta-analysis as the most commonly used measures of mood in other studies. One study reported results for 2 doses of n–3 PUFA (36), and 1 study reported results for 3 doses of n–3 PUFA (15). Different doses are included as separate studies in the meta-analysis.

The pooled standardized difference in means (ie, the pooled effect size) obtained with a fixed-effects model was 0.13 SDs (95% CI: 0.01, 0.25). The pooled standardized difference in means obtained with a random-effects model was 0.50 SDs (95% CI: 0.19, 0.81). Results of the meta-analysis obtained with the use of a fixed-effect model are summarized in **Figure 2**.

Strong evidence of heterogeneity ($I^2 = 79\%$, $P < 0.001$) was observed. To explore this heterogeneity, a funnel plot was drawn and is shown in **Figure 3**. The funnel plot shows evidence of considerable asymmetry.

To explore the heterogeneity further, a sensitivity analysis to assess the effects of one large trial where effects on depressed mood were not a primary outcome measure was conducted by using eleven trials. The excluded trial (29) was large in size and so contributed greatly to the pooled estimate. By using the 11 remaining trials, the pooled standardized difference in means obtained with a fixed-effects model was 0.38 SDs (95% CI: 0.22, 0.54), and the pooled standardized difference in means obtained with a random effects model was 0.57 SDs (95% CI: 0.25, 0.89). Strong evidence of heterogeneity and funnel plot asymmetry remained ($I^2 = 79\%$, $P < 0.001$).

Second, a meta-regression was conducted on all 12 trials to investigate the effect of trial population on the combined estimate. This provided some evidence that the effect was stronger in trials that involved populations with major depression—the difference in the effect size estimates was 0.73 SDs (95% CI: 0.05, 1.41; $P = 0.04$). This explained some of the heterogeneity—I^2 reduced from 79% to 66%, although significant heterogeneity remained ($P < 0.001$). To investigate this further, sensitivity analyses were conducted for each population type. The

TABLE 1
Chronological list of controlled trials investigating effects of n−3 polyunsaturated fatty acids (PUFAs) on depressed mood[1]

Study and year	Participant group	No. of subjects, total (treatment/placebo)	Intervention	Daily dose (n−3 PUFA)	Duration (d)	Treatment status	Outcome measure	Intervention concealed	Blind[2]	ITT analysis
Behan et al, 1990 (25)	Chronic fatigue syndrome	63 (39/24)	EPA + DHA	0.14 g EPA 0.09 g DHA	90	Unclear	4-point Likert scale	No	Yes	No
Warren et al, 1999 (26)	Chronic fatigue syndrome	50 (24/26)	EPA + DHA	0.14 g EPA 0.09 g DHA	90	No existing therapy	BDI	Yes	Yes	No
Stoll et al, 1999 (13)	Bipolar disorder	30 (14/16)	EPA + DHA	6.2 g EPA 3.4 g DHA	112	Adjunctive	HDRS	No	Yes	No
Fenton et al, 2001 (16)	Schizophrenia	87 (43/44)	E-EPA	3.0 g	112	Adjunctive	MADRS	Yes	Yes	No
Keck et al, 2002 (27)	Bipolar disorder	116 (59/57)	E-EPA	6.0 g	120	Adjunctive	IDS-C	No	Yes	No
Nemets et al, 2002 (14)	Unipolar depressive disorder	20 (10/10)	E-EPA	2.0 g	28	Adjunctive	HDRS	Yes	Yes	No
Peet and Horrobin, 2002 (15)	Major depression	70 (17: 1 g/d, 18: 2 g/d, 17: 4 g/d/18)	E-EPA	1, 2, or 4 g	84	Adjunctive	HDRS, MADRS, BDI	No	Yes	Yes
Llorente et al, 2003 (17)	Postpartum depression	99 (44/45)	DHA	≈0.2 g	120	No existing therapy	BDI	Yes	Yes	Yes
Marangell et al, 2003 (28)	Major depression	36 (18/18)	DHA	2.0 g	42	No existing therapy	MADRS, HDRS	No	No	Yes
Ness et al, 2003 (29)	Angina sufferers	452 (229/223)	Fish (or EPA)[3]	Fish[3]	180	No existing therapy	DSP (depression)	No	No	Yes
Su et al, 2003 (30)	Major depression	28 (14/14)	EPA + DHA	4.4 g EPA 2.2 g DHA	56	Adjunctive	HDRS	No	Yes	No
Zanarini and Frankenburg, 2003 (31)	Borderline personality disorder	30 (20/10)	E-EPA	1.0 g	56	Heterogeneous	MADRS	No	Yes	No
Fux et al, 2004 (32)	Obsessive compulsive disorder	11 (11/11)[4]	E-EPA	2.0 g	42	Heterogeneous	HDRS	No	No	Yes
Hirashima et al, 2004 (33)	Bipolar disorder	21 (12/9)	EPA + DHA	5.0–5.2 g EPA 3.0–3.4 g DHA or 1.3 g EPA 0.7 g DHA	28	Heterogeneous	HDRS	No	Yes	Yes
Silvers et al, 2005 (34)	Major depression	77 (40/37)	EPA + DHA	0.6 g EPA 2.4 g DHA	84	Adjunctive	HDRS-SF, BDI	Yes	Yes	Yes
Fontani et al, 2005 (35)	Nonclinical, healthy volunteers	33 (33/33)[4]	EPA + DHA	1.6 g EPA 0.8 g DHA 0.4 g other n−3 fatty acids	35	No existing therapy	POMS (depression)	Yes	Yes	Yes
Frangou et al, 2006 (36)	Bipolar disorder	75 (24: 1 g/d, 25: 2 g/d/26)	E-EPA	1 or 2 g	84	Heterogeneous	HDRS	Yes	Yes	Yes
Nemets et al, 2006 (37)	Major depression (children aged 6–12 y)	28 (13/15)	EPA + DHA	0.38–0.4 g EPA 0.18–0.2 g DHA	112	Heterogeneous	CDRS, CDI, CGI	Yes	Yes	No

[1] ITT, intention-to-treat; EPA, eicosapentaenoic acid; DHA, docosahexaenoic acid; E-EPA, ethyl eicosapentaenoate; LA, linolenic acid; GLA, γ-linolenic acid; BDI, Beck Depression Inventory; HDRS, Hamilton Depression Rating Scale; HDRS-SF, HDRS short form; MADRS, Montgomery-Asberg Depression Rating Scale; DSP, Derogatis Stress Profile; POMS, Profile of Mood States (depression question); CDRS, Children's Depression Rating Scale; CDI, Children's Depression Inventory; CGI, Clinical Global Impression.

[2] Yes = reported; No = not reported or unclear.

[3] The subjects were advised to eat more fatty fish (mackerel, herring, kipper, pilchard, sardine, salmon, or trout) or were given EPA capsules; the recommended dose was not reported.

[4] Within-subjects crossover design.

TABLE 2
Results of controlled trials investigating effects of n−3 polyunsaturated fatty acids (PUFAs) on depressed mood[1]

Study and year	Depression measure	Baseline value	End follow-up	Change	Reported *P* value	Meta-analysis: Effect size θ	Meta-analysis: SE of θ	Meta-analysis: 95% CI
Behan et al, 1990 (25)[2]	4-point Likert scale	n−3 PUFA: 1.4 Placebo: 1.6	n−3 PUFA: 0.6 Placebo: 1.4	n−3 PUFA: 0.8 Placebo: 0.2	< 0.01	—	—	—
Warren et al, 1999 (26)[3]	BDI	n−3 PUFA: 15.0 Placebo: 15.0	n−3 PUFA: 12.0 Placebo: 11.0	n−3 PUFA: 2.5 Placebo: 4.0	0.53	—	—	—
Stoll et al, 1999 (13)[4]	HDRS	n−3 PUFA: 9.5 ± 5.7 Placebo: 12.6 ± 9.1	n−3 PUFA: 4.9 ± 5.3 Placebo: 15.7 ± 9.1	n−3 PUFA: 4.6 Placebo: 3.1	0.002	1.39	0.41	(0.58, 2.20)
Fenton et al, 2001 (16)[4]	MADRS	n−3 PUFA: 8.5 ± 6.6 Placebo: 8.9 ± 5.8	n−3 PUFA: 6.2 ± 4.2 Placebo: 6.6 ± 4.7	n−3 PUFA: 2.3 Placebo: 2.3	0.28	0.09	0.21	(−0.33, 0.51)
Keck et al, 2002 (27)	IDS-C	n−3 PUFA: NR Placebo: NR	n−3 PUFA: NR Placebo: NR	n−3 PUFA: NR Placebo: NR	0.82	—	—	—
Nemets et al, 2002 (14)[4]	HDRS	n−3 PUFA: 24.0 ± 2.9 Placebo: 22.3 ± 2.8	n−3 PUFA: 11.6 ± 6.2 Placebo: 20.0 ± 8.8	n−3 PUFA: 12.4 Placebo: 2.3	< 0.01	1.06	0.48	(0.11, 2.01)
Peet and Horrobin, 2002 (15)[2]	HDRS	1 g/d n−3 PUFA: 19.9 2 g/d n−3 PUFA: 19.6 4 g/d n−3 PUFA: 18.7 Placebo: 20.3	1 g/d n−3 PUFA: 10.0 2 g/d n−3 PUFA: 13.8 4 g/d n−3 PUFA: 12.3 Placebo: 14.2	1 g/d n−3 PUFA: 9.9 2 g/d n−3 PUFA: 5.8 4 g/d n−3 PUFA: 6.4 Placebo: 6.1	0.06	0.60 0.06 0.27	0.35 0.33 0.34	(−0.08, 1.28) (−0.60, 0.71) (−0.39, 0.94)
	MADRS	1 g/d n−3 PUFA: 22.9 2 g/d n−3 PUFA: 20.9 4 g/d n−3 PUFA: 22.6 Placebo: 24.3	1 g/d n−3 PUFA: 11.7 2 g/d n−3 PUFA: 17.9 4 g/d n−3 PUFA: 14.1 Placebo: 18.9	1 g/d n−3 PUFA: 11.2 2 g/d n−3 PUFA: 3.0 4 g/d n−3 PUFA: 8.5 Placebo: 5.4	0.003	—	—	—
	BDI	1 g/d n−3 PUFA: 21.5 2 g/d n−3 PUFA: 22.0 4 g/d n−3 PUFA: 22.6 Placebo: 25.9	1 g/d n−3 PUFA: 9.0 2 g/d n−3 PUFA: 16.3 4 g/d n−3 PUFA: 13.3 Placebo: 19.4	1 g/d n−3 PUFA: 12.5 2 g/d n−3 PUFA: 5.7 4 g/d n−3 PUFA: 9.3 Placebo: 6.5	0.02	—	—	—
Llorente et al, 2003 (17)[4]	BDI	n−3 PUFA: 7.1 ± 4.7 Placebo: 6.5 ± 4.2	n−3 PUFA: 5.8 ± 7.1 Placebo: 4.8 ± 5.9	n−3 PUFA: 1.3 Placebo: 1.7	NR	−0.15	0.21	(−0.57, 0.26)
Marangell et al, 2003 (28)[4]	MADRS	n−3 PUFA: 25.3 ± 5.5 Placebo: 27.2 ± 4.0	n−3 PUFA: 16.2 ± 8.0 Placebo: 21.8 ± 10.5	n−3 PUFA: 9.1 ± 8.3 Placebo: 5.4 ± 9.5	0.23	—	—	—
	HDRS	n−3 PUFA: 23.5 ± 3.1 Placebo: 28.5 ± 4.5	n−3 PUFA: 15.4 ± 8.3 Placebo: 22.7 ± 9.2	n−3 PUFA: 8.1 ± 7.7 Placebo: 5.8 ± 8.6	0.43	0.81	0.35	(0.13, 1.50)
Ness et al, 2003 (29)[4]	DSP (depression)	n−3 PUFA: 53.3 ± 11.3 Placebo: 52.9 ± 9.5	n−3 PUFA: 53.2 ± 10.5 Placebo: 51.6 ± 9.4	n−3 PUFA: unclear Placebo: unclear	NR	−0.20	0.09	(−0.39, −0.02)
Su et al, 2003 (30)[4]	HDRS	n−3 PUFA: 22.5 ± 3.9 Placebo: 22.1 ± 3.9	n−3 PUFA: 8.9 ± 3.7 Placebo: 15.7 ± 3.2	n−3 PUFA: 13.6 Placebo: 6.4	0.001	1.91	0.47	(0.99, 2.82)
Zanarini and Frankenburg, 2003 (31)[4]	MADRS	n−3 PUFA: 17.7 ± 8.4 Placebo: 18.0 ± 3.1	n−3 PUFA: 6.2 ± 4.9 Placebo: 8.0 ± 5.5	n−3 PUFA: 11.5 Placebo: 10.0	0.04	0.34	0.39	(−0.42, 1.11)
Fux et al, 2004 (32)[4]	HDRS	n−3 PUFA: 11.3 ± 7.0 Placebo: 11.3 ± 7.0	n−3 PUFA: NR Placebo: NR	n−3 PUFA: NR Placebo: NR	NR	—	—	—
Hirashima et al, 2004 (33)	HDRS	n−3 PUFA: NR Placebo: NR	n−3 PUFA: NR Placebo: NR	n−3 PUFA: NR Placebo: NR	NR	—	—	—
Silvers et al, 2005 (34)[4]	HDRS-SF	n−3 PUFA: 11.5 ± 0.9 Placebo: 12.4 ± 0.9	n−3 PUFA: 7.0 ± 5.7 Placebo: 5.5 ± 6.2	n−3 PUFA: 0.3 Placebo: 0.6	NR	—	—	—
	BDI	n−3 PUFA: 21.9 ± 3.3 Placebo: 23.3 ± 3.5	n−3 PUFA: 11.8 ± 10.0 Placebo: 9.4 ± 10.6	n−3 PUFA: 0.3 Placebo: 1.5	NR	−0.23	0.23	(−0.68, 0.22)
Fontani et al, 2005 (35)[2,5]	POMS (depression)	n−3 PUFA: 48 Placebo: 48	n−3 PUFA: 45 Placebo: 47	n−3 PUFA: NR Placebo: NR	NR	—	—	—
Frangou et al, 2006 (36)[4]	HDRS	1 g/d n−3 PUFA: 14.7 ± 4.3 2 g/d n−3 PUFA: 14.8 ± 5.6 Placebo: 15.4 ± 5.0	1 g/d n−3 PUFA: 9.2 ± 5.4 2 g/d n−3 PUFA: 9.9 ± 6.6 Placebo: 13.5 ± 6.7	1 g/d n−3 PUFA: NR 2 g/d n−3 PUFA: NR Placebo: NR	0.03	0.69 0.53	0.29 0.28	(0.12, 1.27) (−0.03, 1.09)
Nemets et al, 2006 (37)[4]	CDRS	n−3 PUFA: 71 ± 6 Placebo: 67 ± 11	n−3 PUFA: 32 ± 10 Placebo: 53 ± 13	n−3 PUFA: NR Placebo: NR	0.003	1.74	0.45	(0.85, 2.63)
	CDI	n−3 PUFA: NR Placebo: NR	n−3 PUFA: NR Placebo: NR	n−3 PUFA: NR Placebo: NR	0.04	—	—	—
	CGI	n−3 PUFA: NR Placebo: NR	n−3 PUFA: NR Placebo: NR	n−3 PUFA: NR Placebo: NR	0.001	—	—	—

[1] NR, not reported; BDI, Beck Depression Inventory; HDRS, Hamilton Depression Rating Scale; MADRS, Montgomery-Asberg Depression Rating Scale; IDS-C, Inventory of Depressive Symptomatology; DSP, Derogatis Stress Profile; HDRS-SF, HDRS short form; POMS, Profile of Mood States; CDRS, Children's Depression Rating Scale; CDI, Children's Depression Inventory; CGI, Clinical Global Impression.
[2] Values are means; SDs not reported.
[3] Values are medians.
[4] Values are $\bar{x}$ ± SD.
[5] Values were obtained from graphs.

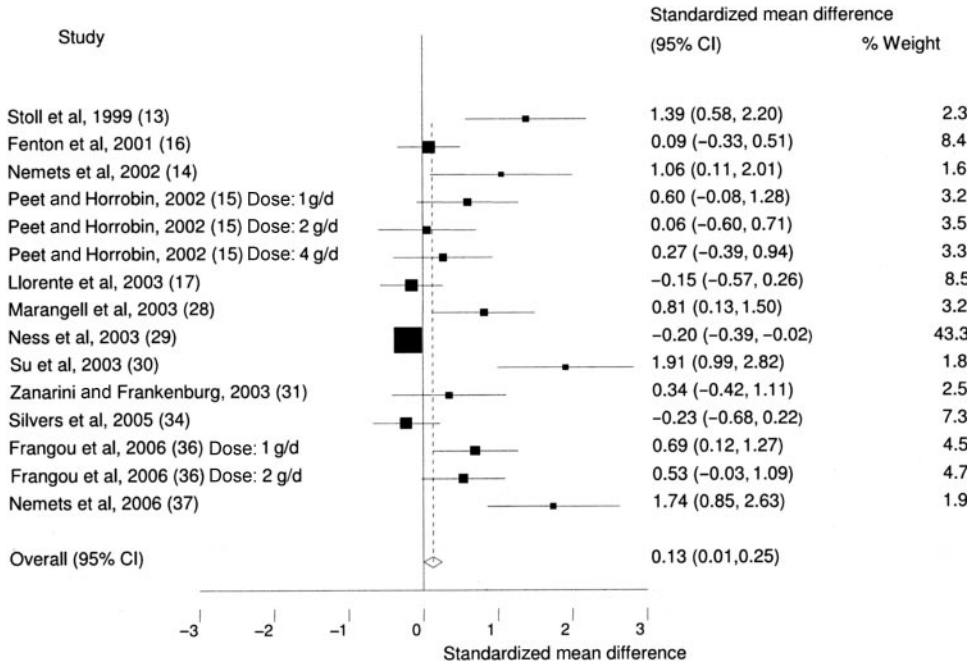

FIGURE 2. Forest plot showing individual and combined effect size estimates and 95% CIs for the 12 trials included in the meta-analysis. ■, The weighting given to the trial in the overall pooled estimate, taking into account the number of participants and the amount of between-study variation (heterogeneity); error bars indicate 95% CIs. —, The combined effect size estimate of 0.13; rhombus, 95% CI (0.01, 0.25).

analysis of effects of n–3 PUFAs on depressed mood in a trial population with major depressive illness diagnoses involved only trials that were conducted in participants who had a diagnosis of unipolar or bipolar depressive illness (13–15, 28, 30, 34, 36, 37). By using these 8 trials, the pooled standardized difference in means obtained with a fixed-effects model was 0.57 SDs (95% CI: 0.37, 0.77), and the pooled standardized difference in means obtained with a random-effects model was 0.73 SDs (95% CI: 0.35, 1.12). Evidence of heterogeneity and funnel plot asymmetry remained ($I^2 = 72\%, P < 0.001$). For the analysis of effects of n–3 PUFAs in trials on persons without major depressive illness (16, 17, 29, 31), the pooled standardized difference in means obtained with a fixed-effects model was −0.13 SDs (95% CI: −0.29, 0.03), and the pooled standardized difference in means obtained with a random-effects model was −0.13 SDs (95% CI: −0.29, 0.02). No evidence of heterogeneity was found

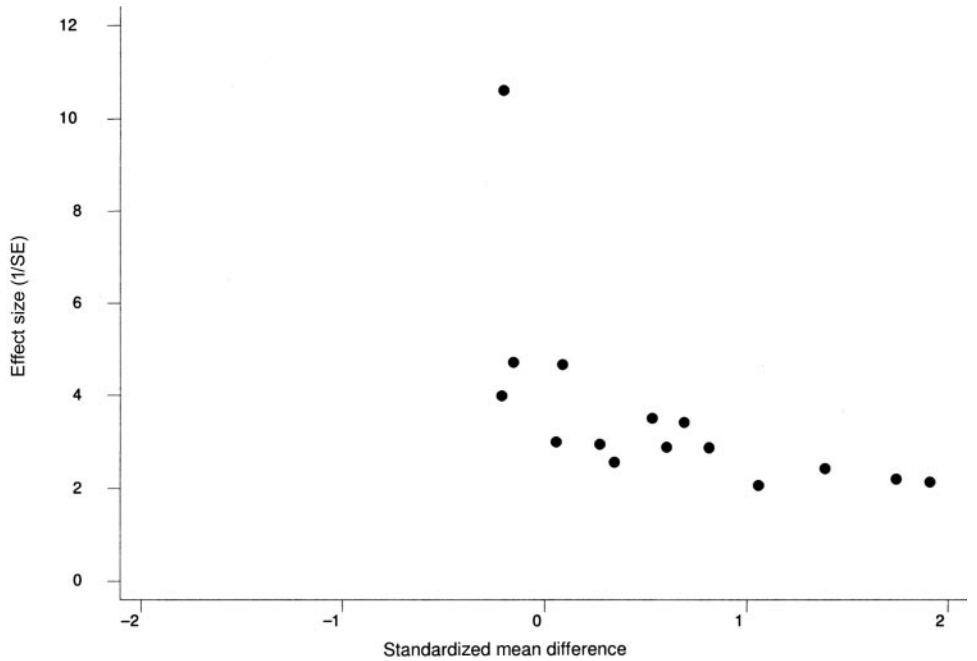

FIGURE 3. Funnel plot of effect size estimates for all individual studies in the meta-analysis.

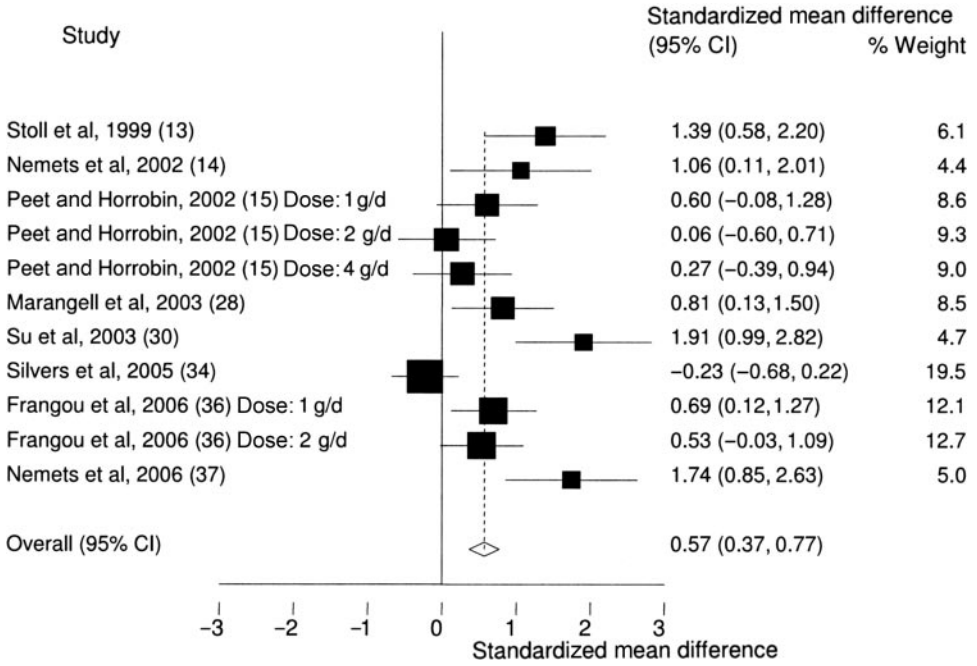

FIGURE 4. Forest plot showing individual and combined effect size estimates and 95% CIs for the 8 trials included in the second sensitivity analysis. ■, The weighting given to the trial in the overall pooled estimate, taking into account the number of participants and the amount of between-study variation (heterogeneity); error bars indicate 95% CIs. —, The combined effect size estimate of 0.57; rhombus, 95% CI (0.37, 0.77).

($I^2 = 3\%$, $P = 0.38$). Results of the sensitivity analysis of the effects of n–3 PUFAs on depressed mood in a trial population with a diagnosis of major depressive illness obtained with a fixed-effects model are summarized in **Figure 4**.

DISCUSSION

Trial evidence that has examined the effect of n–3 PUFAs on depressed mood is very limited. Most trials have been small, of short duration, and have used different combinations of different doses of n–3 PUFAs in varied groups of participants. The substantial differences between trials make combination difficult and potentially misleading.

From the meta-analyses here, the pooled estimate from the fixed-effects model provides little evidence of a beneficial effect of n–3 PUFAs on depressed mood, whereas the combined estimate from the random-effects model suggests a beneficial effect of n–3 PUFAs on depressed mood. There was, however, statistical evidence of substantial heterogeneity and funnel plot asymmetry, which suggests that the combined estimates should be interpreted with considerable caution.

There are several possible causes of heterogeneity and funnel plot asymmetry as seen here (18, 20), the most common source of which is publication bias. If publication bias exists, random-effects models, which place greater weight on small studies, tend to produce estimates of effect size that are biased in the direction of the positive effects of the smaller published studies (18, 20). As publication bias seems likely in these data, the more informative analysis presented in the present review is likely to be the fixed-effects analysis. This analysis suggests that there is little evidence of a beneficial effect of n–3 PUFA for depressed mood.

Of the additional potential sources of heterogeneity that were investigated, the inclusion in the meta-analysis of one large trial where depressed mood was not a primary outcome measure is unlikely to be important. This trial found no effect of n–3 PUFAs on depressed mood compared with placebo and pulled the pooled standardized difference in means toward the null; the meta-analyses conducted excluding this trial resulted in a small beneficial effect of n–3 PUFAs on depressed mood. Heterogeneity, however, was not reduced by the removal of this trial.

The meta-regression, however, provided some evidence that the effect of n–3 PUFA on depressed mood was stronger in trials that involved populations with major depression than in trials conducted on other populations. The meta-analysis performed with only trials that involved populations with a diagnosis of depressive illness (13, 14, 28, 30, 34, 36, 37) showed a beneficial effect of n–3 PUFAs on depressed mood, whereas the meta-analysis performed with the other trials (16, 17, 29, 31) found no evidence of a beneficial effect of n–3 PUFAs on depressed mood. The strong evidence of heterogeneity in the analysis of populations with major depressive illness and the small number of subjects in the analysis of populations without major depressive illness highlight the lack of interpretable evidence. However, these results do suggest that trial population is a modifier of the effects of n–3 PUFAs on depressed mood and that pooling across trials with differing study populations may not be appropriate.

Important additional causes of heterogeneity may also exist in these data. Key potential sources of heterogeneity include measure of depression used (HDRS, HDRS-SF, ADRS, BDI, BDI-II, DSP, or CDRS), the nature of the intervention (n–3 PUFA used, n–3 PUFA dosage, relative proportions of different n–3 PUFAs, and duration of supplementation), characteristics of the trial population (sex, age, and treatment status), and aspects of trial quality. It has been previously suggested that differences in study outcome may be explained by these variables (48). Furthermore, evidence of different effects depending on different measures of depression and different doses of n–3 PUFA supplementation is

available within the studies analyzed here (15, 28, 36). These additional sources of heterogeneity were not investigated here because of the small number of trials available. Only 18 trials were included in the review and only 12 trials included in the meta-analysis; therefore, the ability to explore heterogeneity in the present review was limited. The potential importance of these factors, however, should not be underestimated. The mechanisms underlying any effect of n–3 PUFAs on mood are still far from clearly understood (eg, *see* 49).

Considering the many potential sources of heterogeneity and the small number of trials for which complete data are available, a reliable combined estimate of effects of n–3 PUFAs on depressed mood is difficult to achieve. Additional large, well-conducted trials that have adequate power to detect clinically important differences are required. Power calculations suggest that sample sizes of ≈100 participants per group are necessary to achieve evidence of a clinically meaningful change in depressed mood (3–4 points) as measured by scales such as the HRDS and MADRS. Trial quality is also of paramount importance. Various methodologic aspects of a trial can hugely affect trial outcomes (48). Intervention concealment, the use of blinding, and the use of intention-to-treat analyses were just 3 measures of trial quality assessed here, yet only 3 of 18 studies demonstrated all 3 aspects of trial quality. Recent guidelines for the conduct and reporting of randomized controlled trials (50) will hopefully improve trial quality. The exact trials required are difficult to suggest considering our current lack of knowledge of the mechanisms through which n–3 PUFAs may affect depressed mood. Trials conducted on populations with particularly low n–3 PUFA biochemical status have been previously suggested (48), and, considering the differences found here, trials conducted on populations with diagnoses of major depressive illness may be of value.

It is unfortunate that suitable data (means and SDs of outcome measures) were not available to allow the inclusion of all trials in the meta-analysis. All corresponding authors were contacted, yet relevant data could not be obtained for 6 trials. Two of the 6 trials excluded from the analyses reported positive effects of n–3 PUFAs on depressed mood (25, 35); the other 4 trials reported no beneficial effects (26, 27, 32, 33). It is difficult to assess the impact of these trials. The present review also did not attempt to locate unpublished work or hand search journals to locate additional trials. Other potentially eligible trials, therefore, may have been missed. These trials, however, are likely to be few; the database searches were systematic, thorough, and likely to identify all relevant trials.

In conclusion, little trial evidence is currently available to investigate the effects of n–3 PUFAs on depressed mood, and the evidence that was available is difficult to summarize and evaluate due to the heterogeneous nature of the populations studied and the interventions used. The lack of evidence and the marked heterogeneity between studies highlights the need for well-designed, adequately powered, randomized controlled trials.

We thank Helena Lewis-Smith and Patrick L Burns for their help during data collection and Jonathan AC Sterne (Department of Social Medicine, University of Bristol) for statistical advice. We also thank the corresponding authors of previous articles who provided additional information as required.

All authors were involved in the collection of data for this article and provided intellectual input throughout the project development, analysis, and write-up. The authors had no conflicts of interest.

REFERENCES

1. Horrobin DF. Cardiovascular disease, affective disorders and impaired fatty acid and phosopholipid metabolism. In: Chiu E, Ames D, Katona C, eds. Vascular disease and affective disorders. London, United Kingdom: Martin Dunitz, 2002:332–52.
2. Delion S, Cahlon S, Guilloteau G, et al. Chronic dietary α-linolenic acid deficiency alters dopaminergic and serotinergic neurotransmission in rats. J Nutr 1994;124:266–76.
3. Delion S, Cahlon S, Guilloteau D, et al. α-Linolenic acid deficiency alters age-related changes of dopaminergic and serotinergic neurotransmission in the rat frontal cortex. J Neurochem 1996;66:1582–91.
4. Hibbeln JR, Salem N. Dietary polyunsaturated fatty acids and depression: when cholesterol does not satisfy. Am J Clin Nutr 1995;62:1–9.
5. Hibbeln JR. Fish consumption and major depression. Lancet 1998;351:1213 (letter).
6. Tanskanen A, Hibbeln JR, Tuomilehto J. Fish consumption and depressive symptoms in the general population in Finland. Psych Serv 2001;52:529–31.
7. Silvers KM, Scott KM. Fish consumption and self-reported physical and mental health status. Pub Health Nutr 2002;5:427–31.
8. Timonen M, Horrobin DF, Jokelainen J, Laitinen J, Herva A, Rasanen P. Fish consumption and depression: the Northern Finland 1966 birth cohort study. J Affect Disord 2004;82:447–52.
9. Edwards R, Peet M, Shay J, Horrobin D. Depletion of docosahexaenoic acid in red blood cell membranes of depressive patients. Biochem Soc Trans 1998;26:S142.
10. Peet M, Murphy B, Shay J, Horrobin D. Depletion of omega-3 fatty acid levels in red blood cell membranes of depressive patients. Biol Psych 1998;43:315–9.
11. Adams PB, Lawson S, Sanigorski A, Sinclair AJ. Arachidonic to eicosapentaenoic acid ratio in blood correlates positively with clinical symptoms of depression. Lipids 1996;31:S167–76.
12. Maes M, Christophe A, Delanghe J, et al. Lowered omega-3 polyunsaturated fatty acids in serum phospholipids and cholesteryl esters of depressed patients. Psych Res 1999;85:275–91.
13. Stoll AL, Severus WE, Freeman MP, et al. Omega-3 fatty acids in bipolar disorder—a preliminary double-blind, placebo-controlled trial. Arch Gen Psych 1999;56:407–12.
14. Nemets B, Stahl Z, Belmaker RH. Addition of omega-3 fatty acid to maintenance medication treatment for recurrent unipolar depressive disorder. Am J Psych 2002;159:477–9.
15. Peet MB, Horrobin DF. A dose-ranging study of the effects of ethyl-eicosapentaenoate in patients with ongoing depression despite apparently adequate treatment with standard drugs. Arch Gen Psych 2003;59:913–9.
16. Fenton WS, Dickerson F, Boronow J, Hibbeln JR, Knable M. A placebo-controlled trial of omega-3 fatty acid (ethyl eicosapentaenoic acid) supplementation for residual symptoms and cognitive impairment in schizophrenia. Am J Psych 2001;158:2071–4.
17. Llorente AM, Jensen CL, Voigt RG, et al. Effect of maternal docosahexaenoic acid supplementation on postpartum depression and information processing. Am J Obstet Gynecol 2003;188:1348–53.
18. Sterne JAC, Egger M, Davey Smith G. Investigating and dealing with publication and other biases in meta-analysis. BMJ 2001;323:101–5.
19. Furukawa TA, Barbui C, Cipriani A, Brambilla P, Watanabe N. Imputing missing standard deviations in meta-analyses can provide accurate results. J Clin Epidem 2006;59:7–10.
20. Deeks JJ, Altman DG, Bradbrun MJ. Statistical methods for examining heterogeneity and combining results from several studies in meta-analysis. In: Egger M, Davey Smith G, Altman DG, eds. Systematic reviews in health care: meta-analysis in context. London, United Kingdom: BMJ Publishing Group, 2001:285–312.
21. Egger M, Davey Smith G. Principles of and procedures for systematic reviews. In: Egger M, Davey Smith G, Altman DG, eds. Systematic reviews in health care: meta-analysis in context. London, United Kingdom: BMJ Publishing Group, 2001:23–42.
22. Higgins JPT, Thompson SG. Quantifying heterogeneity in a meta-analysis. Stat Med 2002;21:1539–58.
23. Higgins JPT, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-analyses. BMJ 2004;327:557–60.
24. Sterne JAC, Egger M, Davey Smith G. Investigating and dealing with publication and other biases. In: Egger M, Davey Smith G, Altman DG, eds. Systematic reviews in health care: meta-analysis in context. London, United Kingdom: BMJ Publishing Group, 2001:189–208.

25. Behan PO, Behan WMH, Horrobin D. Effect of high doses of essential fatty acids on the postviral fatigue syndrome. Acta Neurol Scand 1990; 82:209–16.
26. Warren G, McKendrick M, Peet M. The role of essential fatty acids in chronic fatigue syndrome. Acta Neurol Scand 1999;99:112–6.
27. Keck PE, Freeman MP, McElroy SL, et al. A double-blind placebo controlled trial of eicosapentaenoic acid in rapid cycling bipolar disorder. Bipolar Disorders 2002;4(suppl):26–7 (abstr).
28. Marangell LB, Martinez JM, Zboyan HA, Kertz B, Seung Kim HF, Puryear LJ. A double-blind, placebo-controlled study of the omega-3 fatty acid docosahexaenoic acid in the treatment of major depression. Am J Psych 2003;160:996–8.
29. Ness AR, Gallacher JEJ, Bennett PD, et al. Advice to eat fish and mood: a randomised controlled trial in men with angina. Nutr Neurosci 2003; 6:63–5.
30. Su K-P, Huang S-Y, Chiu C-C, Shen WW. Omega-3 fatty acids in major depressive disorder—a preliminary double-blind placebo-controlled trial. Euro Neuropsychopharmacol 2003;13:267–71.
31. Zanarini MC, Frankenburg MD. Omega-3 fatty acid treatment of women with borderline personality disorder: a double-blind, placebo-controlled pilot study. Am J Psych 2003;160:167–9.
32. Fux M, Benjamin J, Nemets B. A placebo-controlled cross-over trial of adjunctive EPA in OCD. J Psych Res 2004;38:323–5.
33. Hirashima F, Parow AM, Stoll AL, et al. Omega-3 fatty acid treatment and T2 whole brain relaxation times in bipolar disorder. Am J Psych 2004;161:1922–4.
34. Silvers KM, Woolley CC, Hamilton FC, Watts PM, Watson RA. Randomised double-blind placebo-controlled trial of fish oil in the treatment of depression. Prost Leuk Ess Fatty Acids 2005;72:211–8.
35. Fontani G, Corradeschi F, Felici A, et al. Blood profiles, body fat and mood state in healthy subjects on different diets supplemented with Omega-3 polyunsaturated fatty acids. Eur J Clin Invest 2005;35:499–507.
36. Frangou S, Lewis M, McCrone P. Efficacy of ethyl-eicosapentaenoic acid in bipolar depression: randomised double-blind placebo-controlled study. Br J Psychiatry 2006;188:46–50.
37. Nemets H, Nemets B, Apter A, Bracha Z, Belmaker RH. Omega-3 treatment of children's depression: a controlled, double-blind pilot study. Am J Psychiatry 2006;163:1098–100.
38. Hamilton M. A rating scale for depression. J Neurol Neurosurg Psych 1960;23:56–62.
39. Montgomery SA, Asberg M. A new depression scale designed to be sensitive to change. Br J Psych 1979;134:382–9.
40. Beck AT, Ward CJ, Mendelson M, Mock J, Erbaugh J. An inventory for measuring depression. Arch Gen Psych 1961;4:53–63.
41. Reynolds W, Kobak K. Hamilton Depression Inventory: a self report version of the Hamilton Depression Rating Scale. Odessa, FL: Professional Manual, Psychological Assessment Resources, 1995.
42. Rush AJ, Gullion CM, Basco MR, Jarrett RB, Trivedi MH. The Inventory of Depressive Symptomatology (IDS): psychometric properties. Psychol Med 1996;26:477–86.
43. Poznanski EO. Cook SC. Carroll BJ. A depression rating scale for children. Pediatrics, 1979;64:442–50.
44. Kovacs M, Beck AT. An empirical-clinical approach toward a definition of children's depression. In: Schulterbrandt JG, Raskin A, eds. Depression in childhood. New York, NY: Raven, 1977.
45. Guy W. ECDEU assessment manual for psychopharmacology: publication ADM 70–338. Washington, DC: US Department of Health, Education and Welfare, 1976:218–22.
46. Derogatis LR. 'The Derogatis Stress Profile (DSP)': quantifications of psychological stress. Adv Psychosom Med 1987;14:30–54.
47. McNair DM, Lorr M, Droppleman LF. Manual of the profile of the mood states. San Diego, CA: Educational and Industrial Testing Service, 1981.
48. Sontrop J, Campbell MK. ω-3 polyunsaturated fatty acids and depression: a review of the evidence and a methodological critique. Preventive Med 2005;42:4–13.
49. Logan AC. Neurobehavioral aspects of omega-3 fatty acids: possible mechanisms and therapeutic value in major depression. Alt Med Rev 2003;8:410–42.
50. Moher D, Schulz KF, Altman D. The CONSORT statement: revised recommendations for improving the quality of reports of parallel-group randomized trials. JAMA 2001;285:1987–91.