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ORIGINAL ARTICLE

Omega-3 fatty acids for the treatment of depression: systematic review and meta-analysis

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We conducted a meta-analysis of randomized, placebo-controlled trials of omega-3 fatty acid (FA) treatment of major depressive disorder (MDD) in order to determine efficacy and to examine sources of heterogeneity between trials. PubMed (1965-May 2010) was searched for randomized, placebo-controlled trials of omega-3 FAs for MDD. Our primary outcome measure was standardized mean difference in a clinical measure of depression severity. In stratified meta-analysis, we examined the effects of trial duration, trial methodological quality, baseline depression severity, diagnostic indication, dose of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) in omega-3 preparations, and whether omega-3 FA was given as monotherapy or augmentation. In 13 randomized, placebo-controlled trials examining the efficacy of omega-3 FAs involving 731 participants, meta-analysis demonstrated no significant benefit of omega-3 FA treatment compared with placebo (standard mean difference (SMD) = 0.11, 95% confidence interval (CI): −0.04, 0.26). Meta-analysis demonstrated significant heterogeneity and publication bias. Nearly all evidence of omega-3 benefit was removed after adjusting for publication bias using the trim-and-fill method (SMD = 0.01, 95% CI: −0.13, 0.15). Secondary analyses suggested a trend toward increased efficacy of omega-3 FAs in trials of lower methodological quality, trials of shorter duration, trials which utilized completers rather than intention-to-treat analysis, and trials in which study participants had greater baseline depression severity. Current published trials suggest a small, non-significant benefit of omega-3 FAs for major depression. Nearly all of the treatment efficacy observed in the published literature may be attributable to publication bias.
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Introduction

Pharmacological and behavioral treatments for major depressive disorder (MDD) show only modest efficacy. For instance, serotonin reuptake inhibitors produce response in fewer than 60%, who complete a full course of serotonin reuptake inhibitor pharmacotherapy.[1] These medications are not always well tolerated; in randomized controlled trials, ~30% of subjects who begin treatment with selective serotonin reuptake inhibitors or tricyclic antidepressants drop out.[2] Thus there is a continued need for the development of both better and better-tolerated pharmacological interventions for MDD.

Fatty acids (FAs) are essential components of cell membranes. Unsaturated FAs have one or more double bonds between carbon atoms; when the double bond is in position 6 the unsaturated FA is called an 'omega-6 FAs' while those with a double bond in position 3 are 'omega-3 FAs', examples of which are eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). In the western diet omega-6 FAs or their precursors (for example, linoleic acid) are much more abundant than omega-3 FAs or their precursors (for example, alpha-linolenic acid). A high omega-6 to omega-3 ratio can alter cell membrane properties and increase production of inflammatory mediators because arachidonic acid, an omega-6 FA found in cell membranes, is the precursor of inflammatory eicosanoids, such as prostaglandins and thromboxanes. By contrast, omega-3 FAs are anti-inflammatory. Therefore, a high dietary omega-6 to omega-3 fatty ratio could promote neuroinflammation. Increased omega-3 FAs concentration in the diet may also act by altering central nervous system cell membrane fluidity and phospholipid composition, which may alter the structure and function of the proteins embedded in it. By this mechanism, increased omega-3 FA concentrations in cell membranes have been shown to affect serotonin and dopamine neurotransmission.[3]

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Evidence from ecological, cross-sectional and case–control studies suggest that fish consumption and omega-3 FA intake may affect the prevalence of MDD. There is a strong negative correlation between fish consumption and national rates of MDD.[4] Cross-sectional studies have demonstrated higher rates of MDD in individuals who rarely consume fish.[5] In case–control studies, individuals with MDD have higher ratio of arachidonic acid to EPA in both cell-membrane cholesteryl esters and serum phospholipids, and significantly higher omega-6/omega-3 ratio than non-depressed controls.[6] Similarly, case–control studies have demonstrated significantly lower omega-3 levels in erythrocyte membranes of depressed patients.[7]

Several double-blind and placebo-controlled trials have studied the efficacy of omega-3 supplementation in adults with MDD. In 2006–2007, three meta-analyses reported a significant benefit of omega-3 FAs for the treatment of depressed mood.[8–10] These meta-analyses suggested a modest effect size of treatment (effect size: 0.13–0.61), a large degree of heterogeneity between studies, and evidence of publication bias in the literature.[8–10] These meta-analyses also included trials that examined depressed mood as an outcome in patients with other primary psychiatric disorders, such as bipolar disorder, schizophrenia, obsessive-compulsive disorder, personality disorders or chronic fatigue syndrome. After these meta-analyses were published, there have been at least nine additional randomized, placebo-controlled trials examining the efficacy of omega-3 FAs for MDD. The addition of these trials more than doubles the sample size of previous meta-analyses and gives us the power to examine the efficacy of omega-3 FAs in MDD specifically. Two recent meta-analyses reported significant treatment benefits of omega-3 FAs in depressed mood consistent with the earlier studies (effect size: 0.10–0.47), but still included patients with other primary psychiatric disorders and failed to adjust for publication bias.[11,12] Trials that report on depressive symptoms as a secondary outcome in other psychiatric illnesses may be particularly prone to publication bias, as authors are less likely to analyze and report secondary measures that are non-significant and therefore non-informative. Furthermore, adding trials in which exclusively included individuals with other primary psychiatric disorders will likely increase heterogeneity between trials.

The goals of this meta-analysis are two-fold: (1) to analyze the efficacy of omega-3 FAs in the treatment of MDD and (2) to examine possible sources of heterogeneity between trials. We specifically hypothesize that possible sources of heterogeneity include (1) omega-3 FA dose, (2) trial duration, (3) diagnostic heterogeneity, (4) the use of omega-3 treatment as monotherapy vs augmentation and (5) baseline depression severity. These sources of heterogeneity represent potentially important differences affecting the efficacy omega-3 FA pharmacotherapy in clinical practice.

Materials and methods

Search strategy

All meta-analytic methods and sensitivity analyses were specified before conducting the meta-analysis but were not registered online. PubMed (1965-May 2010) was searched by two reviewers (JH and MHB) for relevant trials using the search strategy (omega-3 or polyunsaturated FAs or fish oil or EPA or DHA or alpha-linolenic acid or cod liver oil) and (depression or depressive disorder or depressed mood or dysthymia or postpartum depression). The results of the search were further limited to randomized control trials and meta-analyses. The references of eligible trials for this meta-analysis as well as any appropriate review articles in this area were additionally searched for citations of further relevant published and unpublished research. No additional efforts were made to search for unpublished works. There were no language limitations.

Criteria for inclusion of studies in this review

Studies were included in this meta-analysis if they were (1) randomized and placebo-controlled trials examining the efficacy of omega-3 FAs in adults with MDD. Trials were considered randomized when the investigators explicitly represented them as such in the Materials and methods section of their published manuscript. Trials examining the efficacy of omega-3 FAs in treating MDD in subjects with medical comorbidity (that is, cardiac disease and Parkinson's disease) or pregnancy were included in this meta-analysis. Trials were included if they examined the efficacy of omega-3 FAs to target depressive symptoms (as a primary outcome) in patients who may not have received a formal psychiatric diagnosis (that is, in a primary care setting). Trials examining the efficacy of omega-3 FAs in treating depressive symptoms in the context of another primary psychiatric disorder, such as bipolar disorder or schizophrenia, were not included. Trials in which pharmacological agents of known efficacy were started at the same time as omega-3 FAs were also excluded.

Meta-analytic methods

Data extraction was performed by two independently working reviewers (MHB and JH) on specially designed Microsoft Excel spreadsheets. Reviewers collected data on methods, participants, intervention and outcome measurements, and other relevant attributes and results of the studies. Any disagreement between reviewers was resolved through discussion and obtaining more information from the study investigators when possible.

The outcome measure selected from each included trial was the difference in mean improvement between the omega-3 FA and placebo group in a clinical rating scale measuring depression severity over the course of the trial. Preferred rating scales for measuring depression severity were the Hamilton

Depression-Rating Scale (HDRS), either the 9-item short form, 17-item, 21-item or 25-items scales, and the Montgomery Åsberg Depression-Rating Scale.[13–15] When available, HDRS score from each study was used. If the HDRS was not available we used the Montgomery Åsberg Depression-Rating Scale. If neither HDRS nor Montgomery Åsberg Depression-Rating Scale data were available, we used the clinician-rated measure of depression that the investigators identified as their primary outcome. When no clinician-rated measures of depression severity were available from a trial then self-report measures were utilized. When the standard deviation of the mean improvement on placebo or omega-3 FAs was not reported in individual studies, this was imputed based on the standard deviation of reported baseline and endpoint depression severity using Cochrane methodology.[16] The formula $SD(improvement) = \sqrt{(SD(baseline)^2 + SD(endpoint)^2 - 2*r*SD(baseline)*SD(endpoint)}$, where r = correlation coefficient was used to calculate the standard deviation of mean improvement. A correlation coefficient of 0.4 was used for this calculation based on the average of computed correlation coefficients in included studies, where all information on standard deviations was available.

Standard mean difference (SMD) was chosen as the summary statistic for meta-analysis and calculated by pooling the standardized mean improvement of each study using RevMan 5 (Review Manager, version 5.1, Copenhagen: The Nordic Cochrane Center, The Cochrane Collaboration, 2011). SMD was favored over weighted mean difference because rating scales differed across studies. A fixed effects model was chosen for meta-analysis because this method of analysis is favored when there is evidence of publication bias in the literature, as this method gives less weight to small studies. Previous meta-analyses in this area have suggested significant publication bias in this literature.[8,9]

Publication bias was assessed by plotting the effect size against sample size for each trial (funnel plot).[17] We used the rank correlation test in Mix 1.7 (Bax L, Yu LM, Ikeda N, Tsuruta H, Moons KG. Development and validation of MIX: comprehensive free software for meta-analysis of casual research data. *BMC Med Res Methodol*, 2006; 6:50) to statistically test for evidence of the publication bias,[18] and the trim-and-fill method to adjust our analysis for the effects of publication bias on the available data.[18]

Heterogeneity of treatment response was assessed visually from the forest plot of weighted mean differences and relative risk of individual studies. Statistical estimates of heterogeneity were also assessed using the I-square heterogeneity statistic in RevMan.

For secondary analyses we performed several subgroup analyses and meta-regression. For subgroup analyses, trials were stratified based on (1) baseline depression severity (mild, moderate or severe) (2) diagnosis (depression with or without co-morbid medical problems and postpartum depression), (3) whether the omega-3 FA preparation was given as monotherapy for depression or given as an augmentation agent to pharmacological treatment, (4) methodological quality of trials and (5) analysis method (intention-to-treat, modified intention-to-treat or completers). For stratification of trials by baseline depression severity, we used traditional cutoffs for the HDRS-17 (mild: <18, moderate: 18–28, severe: >28).[19] Trials that measured depression severity on scales other than the HDRS-17 were converted to this scale based on previously defined algorithms.[20] Overall methodological quality of trials was assessed using the JADAD Scale.[21,22] Because the subgroup analysis based on study quality was not specified *a priori*, we defined subgroups based on a median split of trial scores on this scale (JADAD = 5 vs JADAD < 5). We used the test for subgroup differences in RevMan to determine whether subgroups reduced overall heterogeneity.[23]

Meta-regression was performed in SPSS 19.0 (SPSS Inc., Chicago, IL, USA) using linear regression. Trials were weighted using the generic inverse variance method. Effect size (SMD) of trials was entered as the dependent variable with the variables of interest being the independent variable. We used meta-regression techniques to examine the association between omega-3 and naturally continuous variables such as (1) trial duration, (2) EPA dose and (3) DHA dose in omega-3 preparations. For our primary analysis examining the efficacy of omega-3 FAs for MDD, we used a significance threshold of $P < 0.05$. For secondary analyses we used a Bonferroni correction and set the threshold at $P < 0.006$ in order to adjust for inflation of false-positive error from our eight secondary analyses.

Sensitivity analyses were conducted to determine the robustness of reviewers' conclusions to methodological assumptions made in conducting this systematic review. We conducted a sensitivity analysis to examine our decision to use a fixed-effects rather than random-effects model for meta-analysis. We also conducted a sensitivity analysis excluding studies that did not require a formal psychiatric diagnosis of MDD to determine if including studies that targeted depressive symptoms without a formal psychiatric diagnosis influenced our findings.

Results

Selection of studies

Our search strategy in PubMED yielded 97 articles for consideration in this review. Studies were excluded because the intervention studied was not omega-3 FAs ($n = 46$), research was not performed in humans ($n = 7$), the design was not a randomized controlled trial ($n = 6$) or subjects were psychiatrically healthy ($n = 12$). Additional randomized controlled trials examining the efficacy of omega-3 FAs for psychiatric conditions were excluded because subjects had bipolar disorder,[24–26] obsessive-compulsive disorder,[27] recurrent self-harm,[28] borderline personality disorder,[29] Alzheimer's disease,[30] or because the trial did not include a placebo control group,[31] examined pediatric depression[32] or involved starting concomitant antidepressant medications at the same time as starting omega-3 supplementation.[33,34]

Characteristics of included studies
Table 1 depicts characteristics of trials included in this meta-analysis. We found 13 separate randomized, double-blind, placebo-controlled trials involving 731 participants that compared omega-3 FAs with placebo for the treatment of depressive disorders. Four studies[35–38] reported a significant benefit of omega-3 FAs in the treatment of depression and nine studies reported no significant difference of omega-3 FAs compared with placebo.[39–47] No studies reported a worsening of depressive symptoms with omega-3 FA therapy compared with placebo.

Efficacy of omega-3 for depression
Meta-analysis showed no significant effect of omega-3 FAs for the treatment of depression (SMD = 0.11, 95% confidence interval (CI): −0.04, 0.26, $P = 0.14$). Figure 1 depicts a forest plot of the effect of omega-3 FAs for the treatment of depression compared with placebo. There was evidence of significant heterogeneity among studies ($\chi^2 = 45.3$, df = 12 ($P < 0.00001$); $I^2 = 73\%$). Sensitivity analysis using a random-effects model (SMD = 0.25, 95% CI: −0.07, 0.56, $P = 0.13$) did not affect the overall findings. Similarly excluding the two trials that did not require a formal psychiatric diagnosis did not affect our overall findings (SMD = 0.07, 95%CI: −0.12, 0.27, $P = 0.45$).

The funnel plot was asymmetric and demonstrated an excess of small positive studies suggesting publication bias in the literature (Figure 2). The rank correlation test for publication bias was statistically significant ($\tau_b = 0.47$, $z = 2.3$, $P = 0.02$). When the trim-and-fill method was used to adjust our results for the potential impact of publication bias, nearly all benefits of omega-3 FAs were eliminated (SMD = 0.01, 95% CI: −0.13, 0.15).

EPA dose
Meta-regression demonstrated no significant difference in efficacy of omega-3 FA preparations based on dose of EPA utilized ($\beta = 0.17 \pm$ standard error = 0.14, 95%CI: (−0.14, 0.47), $P = 0.26$).

DHA dose
Meta-regression demonstrated no significant difference in the efficacy of omega-3 FAs in trials based on dose of DHA utilized (($\beta = -0.25 \pm 0.18$, 95%CI: (−0.64, 0.14), $P = 0.19$).

Omega-3 monotherapy vs augmentation therapy
There was no significant difference in estimates of omega-3 FA efficacy based on whether omega-3 FAs were given as monotherapy or augmentation (test for subgroup differences $\chi^2 = 1.8$, df = 1 ($P = 0.17$); $I^2 = 46\%$). The estimate of effect in trials which omega-3 FAs were given as monotherapy was 0.20 (95%CI: 0, 0.39), whereas the effect estimate was –0.02 (95%CI: –0.26, 0.22) in trials in which concomitant pharmacotherapy was allowed.

Diagnostic indication
There was no significant difference in estimates of omega-3 FA efficacy based on whether trials examined their efficacy in MDD, peripartum MDD or MDD with medical comorbidities (test for subgroup differences: $\chi^2 = 1.9$, df = 2 ($P = 0.39$), $I^2 = 0\%$), and the efficacy was similar for those three subgroups (SMD = 0.06, 95% CI: −0.10, 0.23), (SMD = 0.29, 95% CI: −0.09, 0.67) and (SMD = 0.43, 95% CI: −0.31, 1.17), respectively.

Baseline depression severity
Trials in which participants had moderate depressive symptoms at baseline reported a greater efficacy of omega-3 FAs when compared with trials in which participants were only mildly depressed (test for subgroup differences: $\chi^2 = 11.7$, df = 1 ($P < 0.0006$), $I^2 = 91\%$). The effect size for omega-3 in trials in which participants were at least moderately depressed on average at baseline was 0.42 (95% CI: 0.19, 0.65) compared with trials in which participants had only mild depression on average at baseline (SMD = −0.11, (95% CI: −0.30, 0.09) (see Figure 3).

Trial duration
Meta-regression revealed a strong trend toward shorter duration trials demonstrating a greater efficacy of omega-3 FAs in the treatment of depression ($\beta = -0.11 \pm 0.04$, 95%CI: (−0.20, −0.01), $P = 0.028$, $R^2 = 0.37$) (see Figure 4). However, this association did not reach the strict threshold of statistical significance set forth for secondary analyses in this meta-analysis.

Trial quality
Omega-3 FAs showed greater efficacy in trials judged of lower quality (JADAD = 3 or 4) than in trials judged of higher quality (JADAD = 5; test for subgroup differences: $\chi^2 = 7.2$, df = 1 ($P = 0.007$), $I^2 = 86\%$). The effect size for omega-3 FAs in lower quality trials was 0.47 (95% CI: 0.17 0.76) compared with 0 (95% CI: −0.17, 0.17) in higher quality trials (see Figure 5). Lower quality trials as rated by the JADAD scale generally provided insufficient evidence about the appropriateness of randomization or blinding techniques (or both).

Trial analysis method
There was no significant effect of analysis method on the measured efficacy of omega-3 FAs for depression (test for subgroup differences: $\chi^2 = 9.5$, df = 1 ($P = 0.009$), $I^2 = 79\%$). However, there was a strong trend toward trials relying on completers' analysis (SMD = 0.81, 95% CI: 0.23, 1.38) showing greater efficacy than trials relying on modified intention to treat (ITT) (SMD = 0.35, 95% CI: 0.03, 0.67) and ITT (SMD = −0.01, 95% CI: 0.19, 0.16) analysis (see Figure 6).

Table 1 Characteristics of trials included in the meta-analysis of the efficacy of omega-3 fatty acids for depression

Study	*Year*	*Indicaton*	*Formulation*	*EPA dose (g)*	*DHA dose (g)*	*Study duration (weeks)*	*Mono. V. Aug.*	N	*Rating*	*Depression severity*	*Dropouts (%)*	*Analysis*	*JADAD*	*Result*
Peet[35]	2002	Depression	EPA	1,2 or 4	0	12	Augmentation	70	HAM-D-17	Moderate	10 (14)	ITT	5	+ For 1 g only
Nemets[36]	2002	Depression	EPA	2	0	4	Augmentation	20	HAM-D-24	Moderate	1 (5)	ITT	4	+
Su[38]	2003	Depression	Omega-3	4.4	2.2	8	Augmentation	28	HAM-D-SF	Moderate	6 (21)	Completers	4	+
Marangell[39]	2003	Depression	DHA	0	2	6	Monotherapy	36	HAM-D-28	Moderate	1 (3)	Modified ITT	3	–
Silvers[40]	2005	Depression	Omega-3	0.6	2.4	12	Augmentation	77	HAM-D-SF	Moderate	18 (23)	ITT	5	–
Grenyer[41]	2007	Depression	Omega-3	0.6	2.2	16	Augmentation	83	BDI	Mild	23 (28)	ITT	5	–
Su[37]	2008	Perinatal depression	Omega-3	2.2	1.2	8	Monotherapy	36	HAM-D-21	Moderate	12 (33)	Modified ITT	4	+
Rogers[46]	2008	Mild depression	Omega-3	0.6	0.85	12	Monotherapy	218	DASS	Mild	28 (13)	ITT	5	–
Rees[43]	2008	Perinatal depression	Omega-3	0.4	1.6	6	Monotherapy	26	HAM-D-17	Moderate	5 (19)	ITT	5	–
Freeman[42]	2008	Perinatal depression	Omega-3	1.1	0.8	8	Monotherapy	51	HAM-D-21	Mild	20 (34)	Modified ITT	4	–
da Silva[47]	2008	Depression + PD	Omega-3	0.7	0.5	12	Augmentation	31	MADRS	Mild	2 (7)	Completers	3	+
Mischoulon[45]	2009	Depression	EPA	1	0	8	Monotherapy	35	HAM-D-17	Moderate	11 (31)	Modified ITT	5	–
Lucas[44]	2009	Depression	Omega-3	1.05	0.15	8	Monotherapy	29	HAM-D-21	Mild	3 (10)	ITT	5	–

Abbreviations: BDI, beck depression inventory; DASS, Depression Anxiety Stress Scales; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; HAM-D, Hamilton-rating scale for depression; ITT, intention-to-treat analysis; MADRS, montgomery åsberg depression-rating scale; PD, perinatal depression; QRS, quality-rating scale. All 13 trials included in this meta-analysis were randomized, double-blind and placebo-controlled. All trials were additionally conducted in an outpatient setting relying on referred volunteers. For classification of trials by baseline depression severity we used traditional cutoffs for the HDRS-17 (mild: <18, moderate: 18–28, severe: >28).[19] Trials that measured depression severity on scales other than the HDRS-17 were converted to this scale based on previously defined algorithms.[20]
Modified ITT, ITT analysis where subjects were required to either take one medication dose or make one follow-up visit to be included in the analysis.
+, Positive study, which showed a benefit of omega-3 fatty acids compared with placebo.
–, Negative study, which failed to show benefit of omega-3 fatty acids.
EPA/DHA, ratio of EPA to DHA.

Discussion

Our meta-analysis demonstrates a small, non-significant benefit of omega-3 FAs for the treatment of MDD. There was evidence of publication bias, and statistical adjustment or this bias eliminated nearly all of the observed benefit of omega-3 FAs. This result stands in contrast to many previous meta-analyses that have reported significant benefits of omega-3 FAs in MDD.[8–12]

Several factors are likely responsible for the difference in our results from previous meta-analyses. Since the publication of many previous meta-analyses, eight additional trials examining the efficacy of omega-3 FAs for depression have been published.[8–10] These trials have cumulatively more than tripled the number of MDD participants compared with earlier meta-analyses. The publication of a couple larger trials as well as the relatively recent requirement for public registration of trials and reporting of their primary outcome may be responsible for the decreased evidence of publication bias and treatment effects in later trials (see Figure 2). A greater awareness of the methodological limitations of early omega-3 trials, such as blinding issues caused by a fishy aftertaste present when the active formulations are refluxed may have inflated estimates of efficacy in earlier trials.[48] The adoption of using flavoring, such as orange or peppermint oil or adding a small amount of fish oil to the placebo preparations has increased in recent years.[48] Also, the use of less fishy tasting formulations may have also improved blinding in later trials. To further support this view, trials of lower quality estimated a greater effect size of omega-3 FAs for depression than higher quality studies (see Figure 5). Additionally, trials that did not account for dropouts in their analysis showed a greater effect of omega-3 FAs than trials that employed intention-to-treat analysis.

Another methodological difference compared with other reviews is that this systematic review excluded omega-3 trials of primary psychiatric disorder other than MDD (for example, bipolar disorder, schizophrenia and obsessive-compulsive disorder).[49] These patients have distinct psychiatric conditions as recognized in Diagnostic and Statistical Manual of Mental Disorders, fourth edition or International Classification of Diseases-10, and are often known to be treated with and respond to different pharmacological interventions. In these trials depression was typically a secondary outcome and reporting of secondary outcomes may be particularly prone to publication bias (especially when specific, detailed data are required for meta-analysis). The inclusion of different psychiatric conditions is also likely a source for increased heterogeneity in previous meta-analyses. This increased heterogeneity may occur because improvement in depression symptoms in these trials may be particularly prone to confounding by other factors. For instance, previous meta-analyses have included trials of subjects with schizophrenia.[50,51] A rated improvement in depressive symptoms in a population of schizophrenics could be attributable to (1) improvement in the underlying psychosis that secondarily affects depression measures (for example, a subject's depression rating improves because he is happy, as his hallucinations are less), (2) improvement in some psychotic symptoms may directly lower depression rating scores (for example, improvement in negative symptoms of schizophrenia would lead to an improvement in depression ratings) or (3) improvement could occur in other symptoms commonly experienced by schizophrenics that could affect depression ratings (for example, if omega-3 FA supplementation decreased the side-effects of antipsychotic medications (such as fatigue and emotional blunting) depression ratings would improve). Despite these differences, the updated results of a systematic review which examined improvement in depressive symptoms among this broad population reported fairly similar findings.[49] This systematic review reported a small (effect size (ES) = 0.10) but significant effect of omega-3 FAs, with considerable evidence of publication bias and heterogeneity.[49] A subsequent correspondence suggested that nearly all the treatment effect observed in the literature might be due to publication bias and that claims regarding efficacy of omega-3 given the publication bias were inappropriate.[52] Our meta-analysis serves to reinforce the findings of this previous systematic review and reinforce the interpretation of later critiques with data. We further demonstrate no evidence of a beneficial treatment effect when the data are specific to those with MDD, which is in contrast to this previous meta-analysis.[49]

Our meta-analysis also demonstrated significant heterogeneity between trials. We conducted several secondary analyses to examine possible sources of this heterogeneity. Although, we did not demonstrate a significant overall effect of omega-3 FAs in MDD, our results suggest that they may be effective in patients with more severe depression. Trials where participants were at least moderately depressed on average showed a significant and medium-sized effect of omega-3 treatment. This finding is quite similar to a recent meta-analysis using patient-level data that demonstrated a significantly greater effect of the antidepressants, paroxetine and imipramine in severe depression.[53] On the other hand, trials that included subjects with more severe depression severity also tended to be of lower methodological quality, employ completer rather than intention-to-treat and have smaller numbers of subjects (and thus be more prone to publication bias). We are unsure of whether this significant finding in secondary analysis is emblematic of a significant treatment effect or due to confounding by these other factors.

There were also several other potential sources of heterogeneity between trials that we were not able to investigate in this meta-analysis. Research has suggested the possibility that genetic variation may influence the extent to which individuals require

Study or Subgroup	Weight	Std. Mean Difference IV, Fixed, 95% CI	Year
Nemets B	2.2%	1.47 [0.45, 2.48]	2002
Peet	7.7%	0.28 [−0.26, 0.82]	2002
Marangell	5.0%	0.28 [−0.39, 0.94]	2003
Su–MDD	2.7%	1.04 [0.13, 1.94]	2003
Silvers	11.1%	−0.25 [−0.70, 0.20]	2005
Grenyer	11.4%	−0.69 [−1.13, −0.25]	2007
Su–PD	4.7%	0.93 [0.24, 1.62]	2008
Rogers	31.7%	0.15 [−0.12, 0.41]	2008
Rees	3.6%	0.66 [−0.14, 1.45]	2008
Freeman	7.3%	−0.31 [−0.86, 0.25]	2008
da Silva	4.1%	0.43 [−0.31, 1.17]	2008
Lucas	3.9%	−0.66 [−1.41, 0.10]	2009
Mischoulon	4.6%	0.88 [0.18, 1.58]	2009
Total (95% CI)	**100.0%**	**0.11 [−0.04, 0.26]**	

Heterogeneity: Chi^2 = 45.26, df = 12 (P < 0.00001); I^2 = 73%
Test for overall effect: Z = 1.47 (P = 0.14)

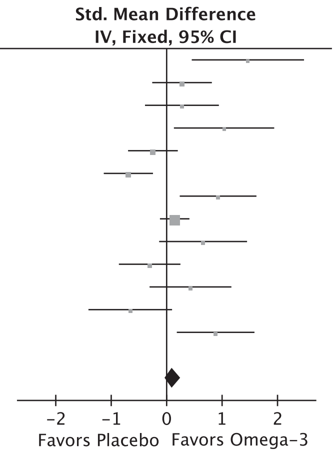

Figure 1 Forest plot of omega-3 FAs for depression. There was no significant effect of omega-3 FAs for major depression compared with placebo. There was significant evidence of heterogeneity between trials. Significant publication bias was also evident using the rank correlation test ($\tau_b = 0.47$, $z = 2.3$, $P = 0.02$).

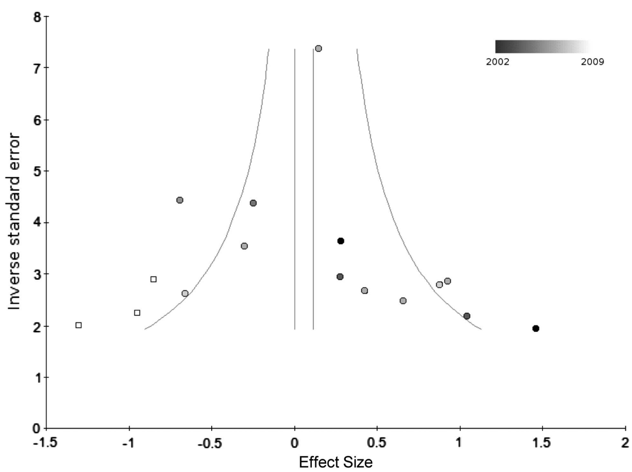

Figure 2 Funnel plot examining publication bias in omega-3 FA trials to treat major depression. This funnel plot depicts the effect size of trials vs their inverse standard error. Published trials are depicted as circles and are shaded from darkest to lightest based on their publication year. White squares represent potentially missing trials that were imputed based on the trim-and-fill method. The red line represents the point estimate for omega-3 treatment effects based on published trials (SMD = 0.11, 95% CI: −0.04, 0.26, $P = 0.14$). The red curves bracket the 95% CI for the expected results of trials given this estimated underlying effect size. The black vertical line represents the new point estimate for the effect size of omega-3 FAs, when publication bias is adjusted for using the trim-and-fill method. (SMD = 0.01, 95% CI: −0.13, 0.15). Abbreviation: SMD, standardized mean difference.

omega-3 FAs.[54] A hypothesis exists that the relationship between omega-3 FAs and depression may be the result of an interaction between low intake of these polyunsaturated FAs in combination with a genetically determined predisposition toward abnormal phospholipid synthesis, which results in a reduced cellular uptake of omega-3 FAs.[55] Reduced cellular uptake of omega-3 FAs has been linked to low activity of FA coenzyme A lipase 4 and/or type IV phospholipase A2, and low-functioning variants for each of these genes have been associated with MDD.[56–58] Also, the ability of omega-3 FAs to suppress production of the inflammatory cytokine, TNF-α may be related to a polymorphism within the lymphotoxin gene.[59] Thus differences between trials regarding the omega-3 FA diet of the samples and genetic heterogeneity could plausibly influence trial results. Few trials measure the omega-3 FA deficiency status of their subjects and even fewer trials assess potentially important sources of genetic heterogeneity. These measures thus represent potentially important sources of heterogeneity that could serve moderators or mediators of treatment effects and deserve further research.[60]

Our meta-analysis has several limitations. The use of patient-level rather than study-level data would have allowed us more powerfully and accurately to examine several potential moderators of treatment effect, such as the influence of baseline depression severity and the use of omega-3 FA supplementation as monotherapy vs as augmentation therapy. The use of patient-level data would have allowed us to also look at other potential moderators of treatment effect such as daily dietary fish consumption or other sources of omega-3 FAs, or mediators such as plasma omega-3 to omega-6 ratio in cell plasma membranes. We suspect that the use of study level data rather than patient-level decreased our power to detect differences in moderators of treatment effect when we were able to examine them. Furthermore, our examination of several secondary factors (in subgroup or meta-regression analysis) leaves open the strong possibility of confounding. For example, a few early trials that were poorly conducted, of short duration, used highly depressed patients and high doses of EPA, generally reported the largest effects of omega-3 FAs. Without patient-level data or a vastly increased population of

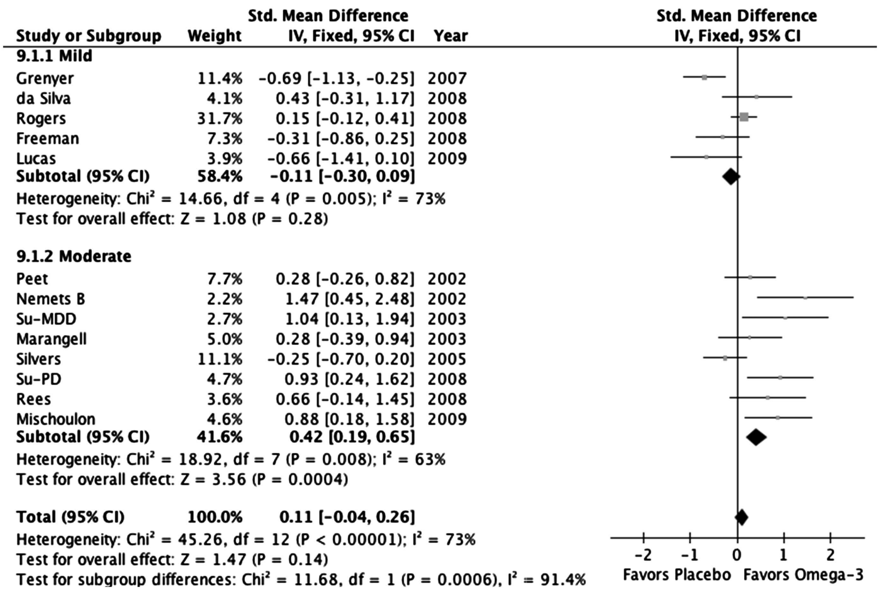

Figure 3 Forest plot of omega-3 FAs for depression stratified by baseline depression severity. Trials in which participants were at least moderately depressed on average before starting treatment, reported a greater efficacy of omega-3 FAs when compared with trials in which participants were only mildly depressed (test for subgroup differences: $\chi^2 = 11.7$, df = 1 ($P < 0.0006$), $I^2 = 91\%$). For stratification of trials by baseline depression severity, we used traditional cutoffs for the HAM-D-17 (mild: <18, moderate: 18–28, severe: >28).[19] Trials that measured initial depression severity on scales other than the HAM-D-17 were converted to this scale based on previously defined algorithms.[20]

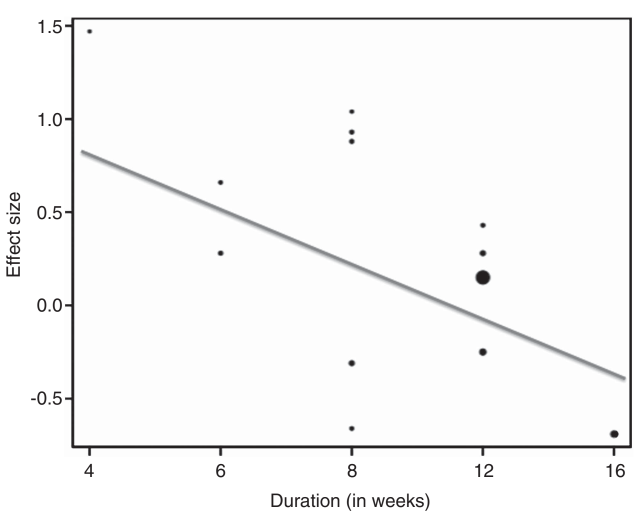

Figure 4 Meta-regression of effect size of omega-3 FAs vs trial duration. Trials of shorter duration tended to show a greater efficacy of omega-3 FA. The size of the circle representing each trial is proportional to its weighing on the overall analysis ($\beta = -0.11 \pm 0.04$, 95%CI: (−0.20, −0.01), $P = 0.028$, $R^2 = 0.37$). Trials were weighed using the generic inverse variance method.

studies, it is impossible to determine which of these factors are most influential.

Our meta-analysis also suggested possible publication bias in the literature. Because publication bias may have inflated our and others estimates of omega-3 efficacy, we decided to adjust for the publication bias using the trim-and-fill method. The detection and adjustment for publication bias is difficult and somewhat controversial when there is a small number of trials.[61] We therefore decided to present our results before and after adjusting for publication bias, and made the primary data for this decision available in Figure 2 so that the reader can make their own decision about validity. There are clearly other reasons that the results of smaller trials may differ systematically from larger trials, such as the use of more severely ill patients, the use of higher doses of omega-3 FAs or greater treatment fidelity, or adherence in smaller studies. However, there are no clear reasons to believe this is the case. Although we examined sources of heterogeneity by conducting secondary analyses, this only explained part of the heterogeneity. We believe that other differences between trials that we were unable to measure, such as differences in (1) participant characteristics—that is, baseline dietary fish consumption and (2) trial methodology—that is, depression scale utilized, adequacy of blinding or tolerability of intervention may be additional sources of heterogeneity.

Despite these limitations our meta-analysis has several important findings. After the inclusion of several recent studies examining the efficacy of omega-3 FAs in MDD, there is no longer significant evidence of efficacy. Meta-analysis also suggests that omega-3 FAs have at most minimal efficacy in treating depression. Further statistical adjustment for publication bias eliminates nearly all of the treatment

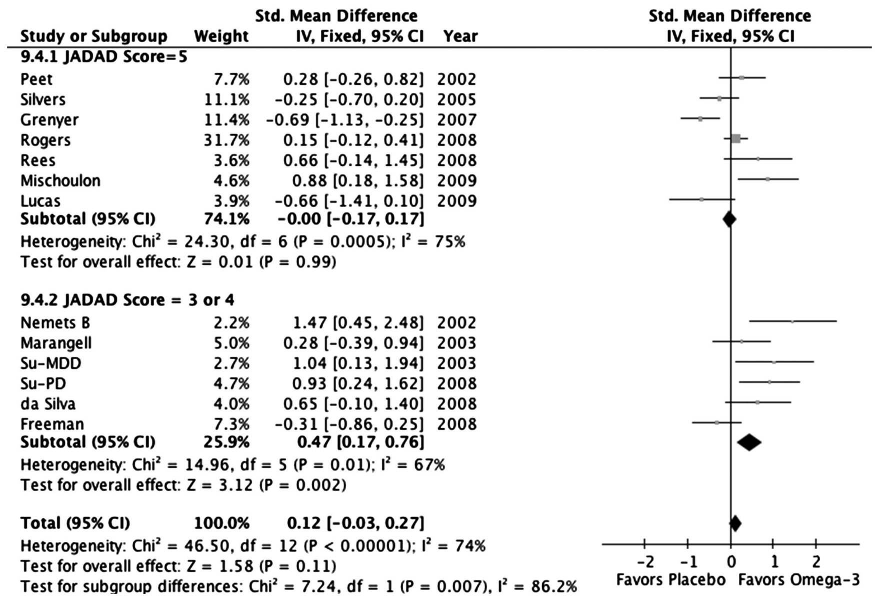

Figure 5 Forest plot of omega-3 FAs for depression stratified by methodological quality of trials. Omega-3 FAs tended to show greater efficacy in trials judged of lower quality (JADAD = 3 or 4) than in trials judged of higher quality (JADAD = 5 test for subgroup differences: $\chi^2 = 7.2$, df = 1 ($P = 0.007$), $I^2 = 86\%$). Clinical trials were stratified based on a median split of scores on the JADAD Scale. The JADAD scale rates trials on presence and appropriateness of randomization and blinding as well as whether the number and reasons for dropouts were described in each trial.

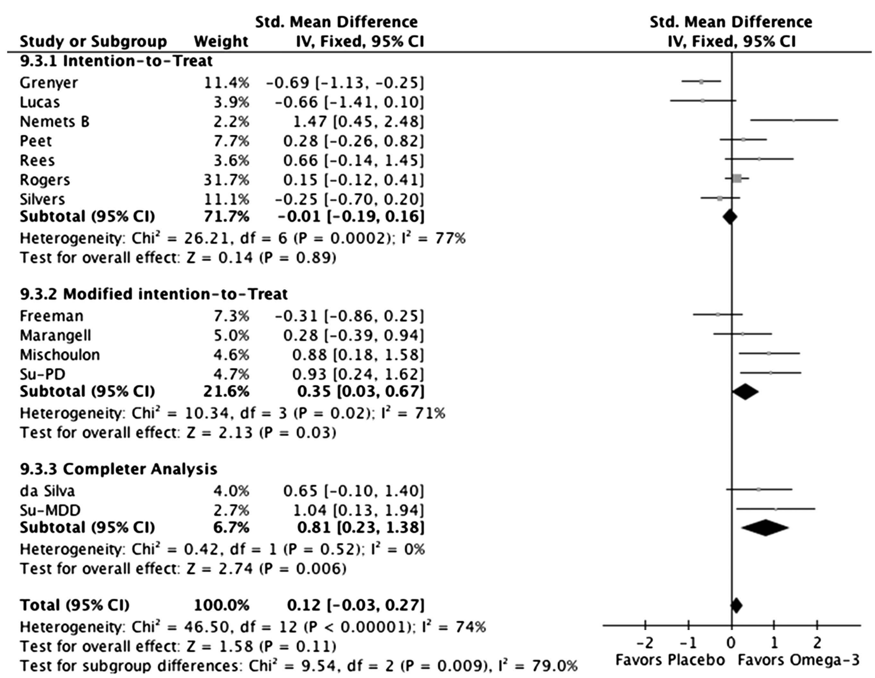

Figure 6 Forest plot of omega-3 FAs for depression stratified by method of accounting for dropouts. Omega-3 FAs tended to shower greater efficacy in trials that utilized completers analysis compared with trials that employed a modified intention-to-treat or an intention-to-treat analysis (Test for subgroup differences: $\chi^2 = 9.5$, df = 1 ($P = 0.009$), $I^2 = 79\%$).

benefits observed in the published literature. These findings are particularly sobering in light of the seven additional double-blind placebo-controlled trials of omega-3 FAs in the treatment of MDD, (including four federally funded trials), being currently conducted according to http://clinicaltrials.gov (accessed 9 December 2010). These ongoing trials plan to enroll over 1000 additional subjects with depression. Although there is still strong evidence based on the epidemiological and cellular literature that omega-3/omega-6 FA balance may have an important role in the pathogenesis of depression, there is limited evidence for omega-3 FA supplementation being an effective acute treatment for it.

Conflict of interest

The authors declare no conflict of interest.

Acknowledgments

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