Review

Omega-3 polyunsaturated fatty acid supplementation and cognition: A systematic review and meta-analysis

Journal of Psychopharmacology
2015, Vol. 29(7) 753–763

DOI: 10.1177/0269881115587958
jop.sagepub.com

Ruth E Cooper, Charlotte Tye, Jonna Kuntsi, Evangelos Vassos* and Philip Asherson*

Abstract

Background: Omega-3 polyunsaturated fatty acids (*n*-3 PUFAs) are promoted as cognitive enhancers with consumption recommended in the general population and those with neurocognitive deficits such as attention deficit hyperactivity disorder (ADHD). However, evidence from randomised placebo-controlled trials is inconclusive.

Aims: This study aimed to conduct a systematic review and meta-analysis examining the effect of *n*-3 PUFA supplementation on cognition in healthy populations and those with ADHD and related disorders (RDs).

Methods: Databases were searched for randomised controlled trials (RCTs) in adults and school-aged children (who were healthy and typically developing (TD) or had ADHD or a related-neurodevelopmental disorder (ADHD+RD) which assessed the effects of *n*-3 PUFA on cognition.

Results: In the 24 included studies *n*-3 PUFA supplementation, in the whole sample and the TD and ADHD+RD subgroup, did not show improvements in any of the cognitive performance measures. In those with low *n*-3 PUFA status, supplementation improved short-term memory.

Conclusions: There is marginal evidence that *n*-3 PUFA supplementation effects cognition in those who are *n*-3 PUFA deficient. However, there is no evidence of an effect in the general population or those with neurodevelopmental disorders. This has important implications given the widespread advertisement and consumption of *n*-3 PUFA; claims of cognitive benefit should be narrowed.

Keywords

Attention deficit hyperactivity disorder, cognition, omega-3, randomised controlled trial, meta-analysis

Introduction

Global spending on omega-3 products is in the billions with consumption recommended in both the general population and those with neurocognitive deficits such as attention deficit hyperactivity disorder (ADHD; Bloch and Qawasmi, 2011), psychosis (Amminger et al., 2010), depression (Su et al., 2014) and autism (Yui et al., 2012). Stimulant medications significantly reduce the symptoms and cognitive impairments in ADHD (Banaschewski et al., 2006; Coghill et al., 2014; Faraone and Buitelaar, 2010). However some individuals elect against such medication due to undesirable side-effects, partial response and questions regarding the long-term efficacy and developmental effects (Dunnick and Hailey, 1995; Leonard et al., 2004; Nasrallah et al., 1986). Omega-3 polyunsaturated fatty acid (*n*-3 PUFA) supplementation is an extensively studied alternative treatment for ADHD, with meta-analyses of behavioural data demonstrating a small but significant effect on ADHD symptom improvement in children (Bloch and Qawasmi, 2011; Sonuga-Barke et al., 2013). It has also been proposed that *n*-3 PUFA supplements are important for the health of the brain and improve cognitive functions (Bryan et al., 2004). However, as yet there has been no systematic evaluation of the available evidence on which to draw any firm conclusions about its efficacy.

Longitudinal and cross sectional studies suggest an association between increased *n*-3 PUFA intake and cognitive function (Aberg et al., 2009; Bryan et al., 2004; Hibbeln et al., 2007). One of the main explanations proposed is based on the high lipid cell membrane composition, maintenance of which may be vital for the optimal development and function of the brain and nervous system (Bryan et al., 2004). However, randomised controlled trials (RCTs) in typically developing (TD) participants and those with ADHD and related neurodevelopmental disorders, have instead yielded mixed results. Benefits of *n*-3 PUFA supplementation on cognitive performance have been reported in healthy adults (Stonehouse et al., 2013) and children with ADHD (Sinn et al., 2008) and developmental coordination disorder (DCD) (Richardson and Montgomery, 2005). Yet a number of other studies in these populations have failed to find an effect (Jackson et al., 2012; Kairaluoma et al., 2009; Milte et al., 2012; Osendarp et al., 2007)

King's College London, MRC Social, Genetic and Developmental Psychiatry Centre, Institute of Psychiatry, Psychology and Neuroscience, London London, UK

*Co-senior authors.

Corresponding author:
Ruth E Cooper, King's College London, MRC Social, Genetic and Developmental Psychiatry Centre, Institute of Psychiatry, Psychology and Neuroscience, London SE5 8AF, UK.
Email: ruth.cooper@kcl.ac.uk

Given the global market for omega-3 products it is of public importance that there is a more conclusive picture as to whether *n*-3 PUFA supplementation improves cognitive performance. We therefore conducted a systematic review and meta-analysis of randomised placebo-controlled trials which examined the effect of *n*-3 PUFA supplementation on cognitive performance in healthy populations and those with ADHD and related neurodevelopmental disorders.

Methods

We conducted a systematic review and meta-analysis in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines (Moher et al., 2009).

Eligibility criteria and data extraction

Studies were included if: (a) they were randomised double-blind placebo-controlled trials of *n*-3 PUFA supplementation including docosahexaenoic acid (DHA), eicosapentaenoic acid (EPA) and alpha-linoleic acid (ALA). Trials supplementing with ALA alone were excluded as ALA is thought to have a limited impact on cognition compared to EPA and DHA (Kalmijn et al., 2004) and humans have a limited capacity to synthesise EPA and DHA from ALA (Goyens et al., 2005); (b) participants were school-aged children (4–12 years), adolescents (13–17 years) or adults (18–60 years) who were either healthy (TD group) or had a diagnosis of ADHD or high levels of ADHD symptoms or related neurodevelopmental traits such as DCD or dyslexia (ADHD+RD group); and (c) the study measured cognitive performance defined as (one or more measure of): intelligence quotient (IQ), inhibition, attention (omission errors), working memory, short-term memory, reading, spelling, mean reaction time and reaction time variability (see Supplementary Material, Table S1 for details). There were no language restrictions on trial eligibility.

The databases Ovid Medline (1946–September, week 2, 2014), Embase (1974–2014, week 37) and Psychinfo (1806–September, week 3, 2014) were searched. References of eligible trials and appropriate reviews were searched for additional citations. Unpublished or ongoing trials were searched on the ClinicalTrials.gov website and authors contacted to request relevant data. The search was updated in November 2014. The search terms used are listed in Supplementary Material, Table S2.

Risk of bias to determine study quality was assessed independently by two authors (REC and CT) according to PRISMA guidelines and the Cochrane Handbook of Systematic Reviews (Higgins and Green, 2011) (Supplementary Material, Table S3 and S4). Decision to include was based on risk of bias which was classed as low, unclear or high. Unresolved classification of studies was arbitrated by PA.

Data extraction was performed by REC and checked by a research assistant. The main outcome measures were the mean and standard deviation (SD) of the pre and post treatment cognitive performance measures for active and placebo arms, with intent to treat (ITT) analysis preferentially reported. Additional measures investigated included participant characteristics, study design and the supplement type and dose. If multiple treatment arms were present, only those supplementing with *n*-3 PUFA or placebo were included. With regard to missing data, we contacted authors. Missing data that remained unavailable was not imputed.

Cognitive performance measures

Nine domains of cognitive performance, previously found to be impaired in ADHD and related disorders (Doyle et al., 2005; Frazier et al., 2004; Kuntsi et al., 2009; Willcutt et al., 2010) were measured in these studies and included in this meta-analysis (see Figure 1). Examples of the main measures and tasks were as follows: IQ measured using the Wechsler Intelligence Scale for Children (WISC; Wechsler, 1991); commission errors (the inability to withhold a pre-potent response) on computerised tasks for inhibition (e.g. continuous performance tasks); omission errors (failing to respond when a response is required) on computerised attention tasks (e.g. test of variables of attention (TOVA) (Greenberg and Kindschi, 1996)) for attention (omission errors); digit span backwards (recalling a string of numbers backwards) for working memory; immediate or delayed word recall for short term memory; reading and spelling using subtests of the Wide Range Achievement Test (WRAT; Wilkinson and Roberts, 2006); mean reaction time (speed of responding) and reaction time variability (the variability in the speed of responding) during attention tasks (e.g. TOVA) (see Supplementary Material, Table S1 for a detailed list of cognitive measures).

Statistical analyses

Analyses were carried out in STATA (StataCorp, 2009) on the whole sample, the TD and ADHD+RD subgroups separately (with a further analysis of adults and children separately in the TD group) and then for the secondary subgroup analysis (see subgroup analysis section). Where a study contained two active groups which were both eligible for inclusion (for example when the active groups differed in the dose of *n*-3 PUFA), they were combined (with the method presented in the *Cochrane Handbook*: section 16.5.4; Higgins and Green, 2011). Effect sizes were estimated as the standardised mean difference (SMD); calculated as the mean pre-to post-treatment change, minus the mean pre-to post-placebo group change, divided by the pooled pre-test standard deviation (SD) with a bias adjustment (Morris, 2007). Effect sizes were classified according to Cohen's *d* (0.2=small, 0.5=medium, 0.8=large; Cohen, 1988). Where SD was not provided, it was calculated from sample size, *p*-values, *t*-values, standard error (SE) or 95% confidence intervals (CIs). For individual studies that contributed multiple assessments for one cognitive domain, a single SMD was derived from a meta-analysis of these assessments (see Supplementary Material, Table S1) hence an individual study was counted only once per cognitive domain. Cross-over trials were treated as parallel group trials using the pre-cross-over data, because insufficient data were provided to permit analysis of within-individual change (e.g. no correlations of scores between conditions). This approach is considered conservative (studies are under-rather than over-weighted) and is equivalent to setting the between-condition correlation to zero (Elbourne et al., 2002). SMDs in each domain were combined using the inverse-variance method where the reciprocal of their variance is used to weight the SMD

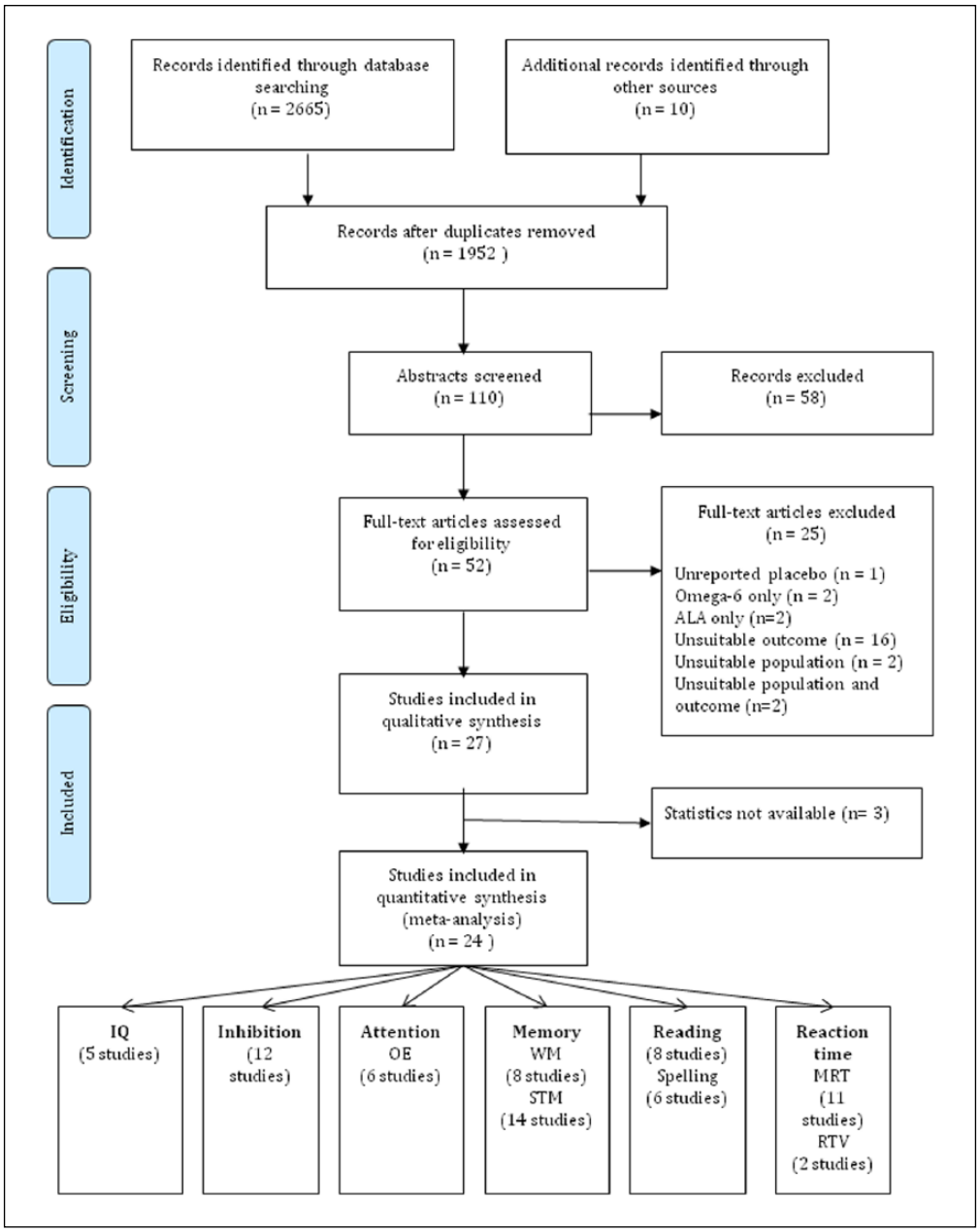


Figure 1. Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) flow diagram.
ALA: alpha-linoleic acid; IQ: intelligence quotient; MRT: mean reaction time; OE: omission error; RTV: reaction time variability; STM: short term memory; WM: working memory.

from each trial before being combined to give an overall estimate (Higgins and Green, 2011). Given the between-study heterogeneity in terms of study design, participant characteristics and outcome measures, we chose a priori to use random effects models (Field and Gillett, 2010). When setting the significance level, we corrected for nine domains of cognition (Bonferroni correction set at $p<0.006$) despite the fact that the primary analysis was performed in the total sample and also separately for the ADHD+RD and TD groups (i.e. more than nine statistical tests were conducted), because the cognitive tests are highly correlated. The above p-value (0.006) was considered indicative and not evidence of association for the post-hoc analyses. A nominal level of significance was set at $p<0.05$. The I^2 statistic assessed heterogeneity between studies. Publication bias was assessed using the Egger regression asymmetry test (and inspection of the regression asymmetry plot) and the Begg adjusted rank correlation test. Meta-regression was used to examine the association between treatment effect and (a) trial duration and (b) dose of EPA and DHA. Four studies contained two active groups (Jackson et al., 2012; Kennedy et al., 2009; McNamara et al., 2010; Milte et al., 2012), therefore the average dose of EPA and DHA was taken across the two groups for the meta-regression and for the 'adequate EPA' subgroup analysis (see 'subgroup analyses' section).

Subgroup analyses

1. Strict inclusion: all studies that met our inclusion criteria were included in the primary analysis (as above). As two studies used supplementation with carnosine (Kairaluoma et al., 2009) or vitamins (Kirby et al., 2010) in addition to *n*-3 PUFA we performed subgroup analyses excluding these two studies.
2. PUFA deficient: it is proposed that only participants who are deficient in *n*-3 PUFA will benefit from treatment. The analysis was therefore run in four studies that supplemented: children of low socio-economic status who had low fish intake (defined in the paper as 'virtually no intake of fatty fish and a very low intake of lean fish,' Dalton et al., 2009, section 2.1), adults with low *n*-3 PUFA intake (less than ~200 mg EPA+DHA/wk, Stonehouse et al., 2013), malnourished children (53% consumed <1 portion fish a week, 39% one portion a week and 8% ⩾2 portions a week, Portillo-Reyes et al., 2014) and children with ADHD who were deficient in *n*-3 PUFA (participants were selected with thirst/skin problems indicative of *n*-3 PUFA deficiency, blood analysis showed these participants to have significantly lower *n*-3 PUFA compared with a TD control group, Stevens et al., 2003).
3. High quality: quality appraisal demonstrated the majority of studies to have design errors therefore the analysis was re-run in the eight studies whose overall risk score was low (and were therefore deemed high quality) (Supplementary Material, Table S3 and S4) (Jackson et al., 2012; Kairaluoma et al., 2009; Karr et al., 2012; Kennedy et al., 2009; Richardson and Montgomery, 2005; Richardson et al., 2012; Stonehouse et al., 2013; Vaisman et al., 2008).
4. Cognitive impairment: heterogeneity in cognitive impairments across study populations may reduce the effect size of treatment response. The analysis was run in four studies which included those with more homogenous cognitive deficits. Milte et al. (2012) tested children with ADHD whose literary performance was behind their year level at school. Vaisman et al. (2008) tested children with ADHD who also performed poorly on a continuous performance test. Richardson et al. (2012) tested a sub-group of the poorest readers (<20th centile from the total sample) and Kairaluoma et al. (2009) tested children with dyslexia.
5. Adequate EPA: a significant association between dose of EPA (but not DHA) and improvement in ADHD symptoms has previously been found (Bloch and Qawasmi, 2011). Given this, it has been suggested that EPA may be more active than DHA in terms of its effect on brain and behaviour. The analysis was therefore run in the 14 studies which supplemented participants with >100 mg EPA (this cut-off was estimated from Figure 3 in Bloch and Qawasmi's paper) (Antypa et al., 2009; Gustafsson et al., 2010; Hamazaki et al., 1996; Jackson et al., 2012; Kairaluoma et al., 2009; Karr et al., 2012; Milte et al., 2012; Parletta et al., 2013; Portillo-Reyes et al., 2014; Richardson and Montgomery, 2005; Sinn et al., 2008; Stonehouse et al., 2013; Vaisman et al., 2008; Widenhorn-Müller et al., 2014).

Results

Selection of studies

The search strategy (conducted by REC) identified 1952 publications. Of these, 110 relevant abstracts were screened, of which 58 were excluded because the studies were not an RCT (*n*=34), or they used an unsuitable outcome (e.g. looked only at treatment effects on PUFA blood levels) (*n*=10), population (*n*=12), supplement (*n*=1), or study design (*n*=1). Fifty-two full text articles were subsequently quality appraised (by REC and CT) and 25 excluded because of failure to report the placebo group (*n*=1), supplementation with omega-6 (*n*=2) or ALA (*n*=2) only, use of unsuitable outcome measures (*n*=16) (e.g. only measured behavioural outcomes), unsuitable population (*n*=2) or unsuitable outcome and population (*n*=2) (Supplementary Material, Table S5 lists the excluded studies). Of the 27 trials suitable for inclusion, after writing to the authors of studies with missing data, the statistical information required for meta-analysis was available for 24 studies, which made up the final dataset used in the meta-analysis (Figure 1 and Supplementary Material, Table S6).

Quality and characteristics of studies included in qualitative synthesis

Randomisation was explicitly described in 20 studies and allocation concealment in 17 studies. In the remainder this was absent or unclear. All studies were double blind apart from one, where the chief investigator was unblinded (although did not collect/analyse data) (Dalton et al., 2009). Inadequate allocation concealment in two studies meant participants were aware they were in different groups (Baumgartner et al., 2012; Dalton et al., 2009). Above-chance guessing (70%) of group allocation occurred in another study (Milte et al., 2012). Drop-outs (*n*=5/25) occurred only in the placebo group in one study (Portillo-Reyes et al., 2014). Reasons for drop-outs were not given in three studies (Benton et al., 2013; Kirby et al., 2010; Ryan and Nelson, 2008) despite one having more than double the amount of drop-outs in the active group (Ryan and Nelson, 2008). In one study the distribution of drop-outs between the placebo and active groups was not given (Gustafsson et al., 2010). In one study the *n*-3 PUFA supplements were taken only four days per week (Baumgartner et al., 2012). In two studies the active groups took supplementation with carnosine or vitamins in addition to *n*-3 PUFA (Kairaluoma et al., 2009; Kirby et al., 2010) (Supplementary Material, Table S3 and S4). Study characteristics are detailed in Supplementary Material, Table S7.

Quantitative meta-analysis

Of the 27 studies included in the qualitative synthesis, pre-and post-treatment means and SDs were not available for three studies (Hirayama et al., 2004; Long and Benton, 2013; Ryan and Nelson, 2008) therefore 24 studies were included in the meta-analysis. Omega-3 PUFA supplementation had no significant effect on any of the nine domains of cognitive performance in either the whole sample or the ADHD+RD or TD group (when analysed as a whole and by adults and children) separately. An effect on working memory in the ADHD+RD group approached significance (three studies, *n*=506) (SMD=0.23; 95% CI: –0.001–0.46, z=1.95, p=0.05) with no heterogeneity (x^2=3.03, I^2=33.9%,

Table 1. Main effects of meta-analysis for the whole sample (combined typically developing (TD), attention deficit hyperactivity disorder (ADHD)+related disorder (RD) group).

Domain	*n* studies	*n* participants	SMD	95% CI	Heterogeneity	
					p	I^2 (%)
IQ	5	434	0.14	−0.07–0.35	0.28	20.9
Inhibition	12	809	−0.04	−0.22–0.14	0.08	38.7
Attention (omission errors)	6	321	−0.13	−0.33–0.07	0.96	0.0
Memory (working memory)	8	1308	0.09	−0.01–0.18	0.40	3.9
Memory (short-term memory)	14	1914	0.07	−0.01–0.15	0.15	29.0
Reading	8	1579	0.02	−0.06–0.09	0.62	0.0
Spelling	6	1167	0.03	−0.09–0.15	0.39	5.0
Reaction time (mean reaction time)	11	1035	−0.002	−0.12–0.12	0.33	12.5
Reaction time (reaction time variability)	2	91	0.29	−0.70–1.28	0.02[a]	82.0

[a]Significant at $p<0.05$.

p=0.22). Main effects from the meta-analysis are summarised in Table 1 and Figure 2 and a detailed description of results is available in Supplementary Material, Section S1.

In the subgroup of those who were *n*-3 PUFA deficient a small treatment effect was found for short-term memory (three studies, n=331, SMD=0.26; 95% CI: 0.09–0.43, z=3.02, p=0.003) with no heterogeneity (x^2=2.67, I^2 =25.1%, p=0.26). In those who met strict inclusion criteria, a small treatment effect emerged for working memory after exclusion of one study which supplemented with vitamins (Kirby et al., 2010) (seven studies, n=960, SMD=0.15; 95% CI: 0.03–0.27, z=2.48, p=0.01) with no heterogeneity (x^2=4.18, I^2=0.0%, p=0.65). In studies that supplemented with adequate EPA a small treatment effect emerged for working memory (five studies, n=510, SMD=0.19; 95% CI: 0.04–0.34, z=2.44, p=0.02) with no heterogeneity (x^2=3.52, I^2=0.0%, p=0.47). Although the latter two treatment effects did not withstand correction for multiple testing (adjusted $p<0.006$). No other significant effects were found. Supplementary Material, Table S8 details the results of the subgroup analysis.

Significant heterogeneity was present in the TD group for IQ (x^2=4.12, I^2=75.8%, p=0.04) and short-term memory (x^2=17.66, I^2=49.0%, p=0.04). In the TD-child sample for inhibition (x^2=3.92, I^2=74.5%, p=0.05), in the TD-adult sample for mean reaction time (x^2=8.34, I^2=64.0%, p=0.04) and in ADHD+RD participants for reaction time variability (x^2=5.54, I^2=82.0%, p=0.02). In the sub-group analyses heterogeneity was found in those who were PUFA deficient for inhibition (x^2=8.16, I^2=75.5%, p=0.02) and mean reaction time (x^2=5.00, I^2=79.3%, p=0.03). Meta-regression found no effect of trial duration or dose of EPA or DHA on any of the eight domains of cognitive performance (there were not enough studies to examine this for reaction time variability (RTV)).

There was evidence of publication bias in working memory (Egger test only) (β=1.87, SE=0.58, t=3.23, p=0.02, 95% CI: 0.45–3.29) but not in any of the other eight domains of cognitive performance.

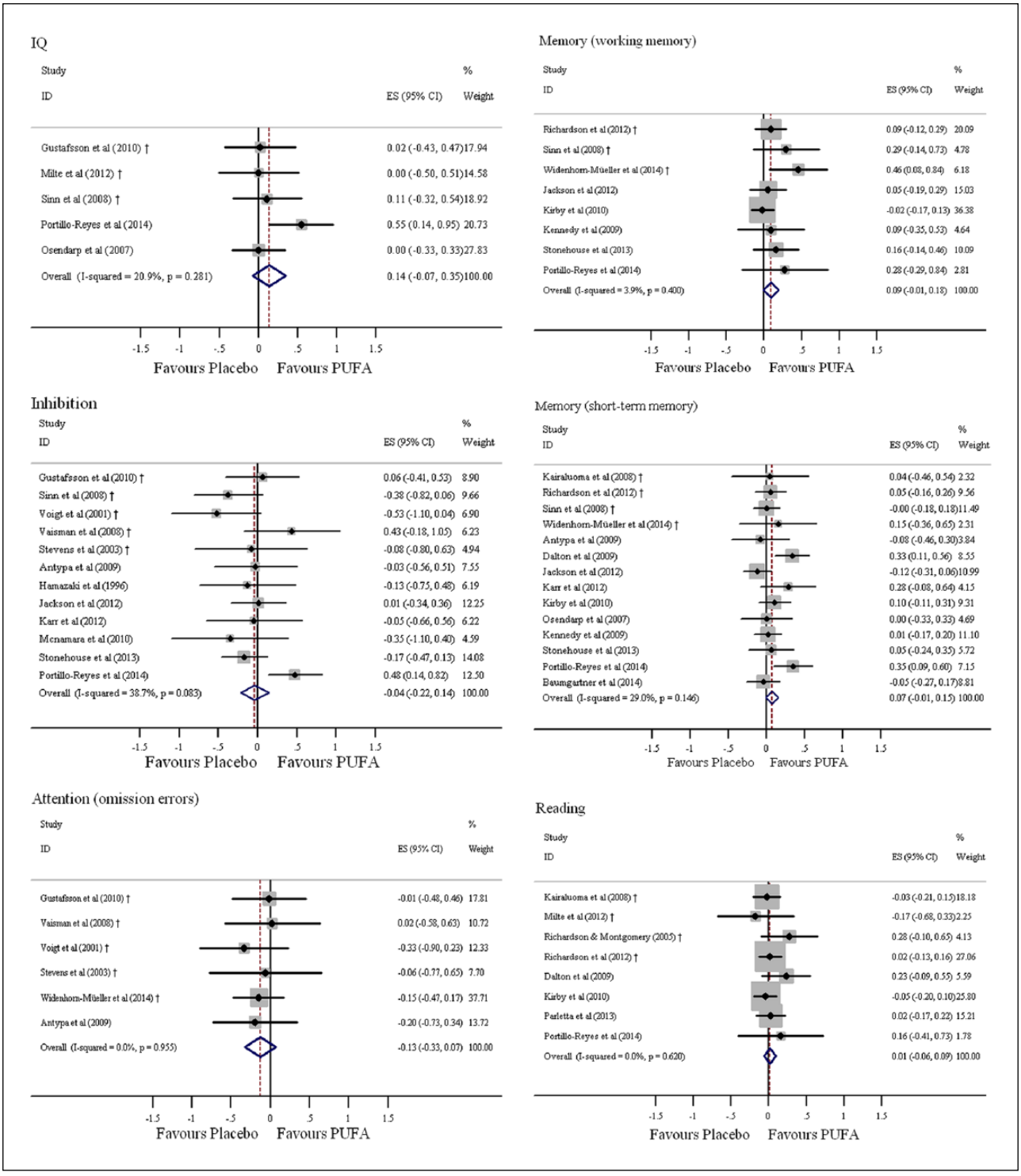
Discussion

This systematic review and meta-analyses examined the efficacy of *n*-3 PUFA supplementation on cognitive performance measures in school aged children and adults who were typically developing (TD) or had ADHD or a related neurodevelopmental disorder (ADHD+RD). We did not find an effect of *n*-3 PUFA supplementation on cognition in either the whole sample or the TD (analysed as a whole and by adults and children separately) or the ADHD+RD group when analysed separately. In the subgroup analyses a small treatment effect emerged for short-term memory in those with low *n*-3 PUFA and for working memory, after removal of a study which supplemented with vitamins (Kirby et al., 2010) and in those studies that supplemented with adequate EPA. However, both the effects on working memory were only nominally significant and were driven by the outcome of one cognitive measure in a small sample of 61 children with ADHD (Widenhorn-Müller et al., 2014).

There was no evidence of heterogeneity in the whole sample. Nominally significant heterogeneity was found in a number of sub-analyses (TD, ADHD+RD, TD-adult, TD-child, PUFA-deficient and high quality studies) this is most likely due to the smaller number of studies included in these analyses. Meta-regression found no effect of trial duration or EPA or DHA dose across any of the eight domains of cognitive performance (there were not enough studies to examine this for RTV). Evidence of publication bias was found only for working memory. We conclude on the basis of these data that there is no evidence of an effect of *n*-3 PUFA supplementation on cognitive performance in typically developing individuals or those with ADHD and related disorders. There is marginal evidence of benefit in those who are *n*-3 PUFA deficient. Evidence for those that met strict inclusion criteria or that supplemented with adequate EPA was much weaker.

A small improvement (which withstood correction for multiple testing) in short-term memory was found across four studies (in TD and ADHD+RD populations) which supplemented those with low *n*-3 PUFA (Dalton et al., 2009; Portillo-Reyes et al., 2014; Stevens et al., 2003; Stonehouse et al., 2013). Results from one study in malnourished children also found improvements in IQ following supplementation (Portillo-Reyes et al., 2014). This is in line with the suggestion that treatment effects on cognitive performance may occur only in those with low *n*-3 PUFA levels at baseline. However only four studies could be included in this subgroup (Dalton et al., 2009; Portillo-Reyes

et al., 2014; Stevens et al., 2003; Stonehouse et al., 2013) and whilst three of them measured PUFA-blood levels, only one of these examined blood-PUFA deficiency. Stevens et al., (2003) found reduced *n*-3 PUFA status in the ADHD study participants compared to TD controls. Therefore it cannot be certain that the other study participants were *n*-3 PUFA deficient. The subgroup analysis on those with low *n*-3 PUFA status found treatment effects in only one of the five cognitive performance domains. Therefore, although promising, further trials are needed before drawing any firm conclusions.



(Continued)

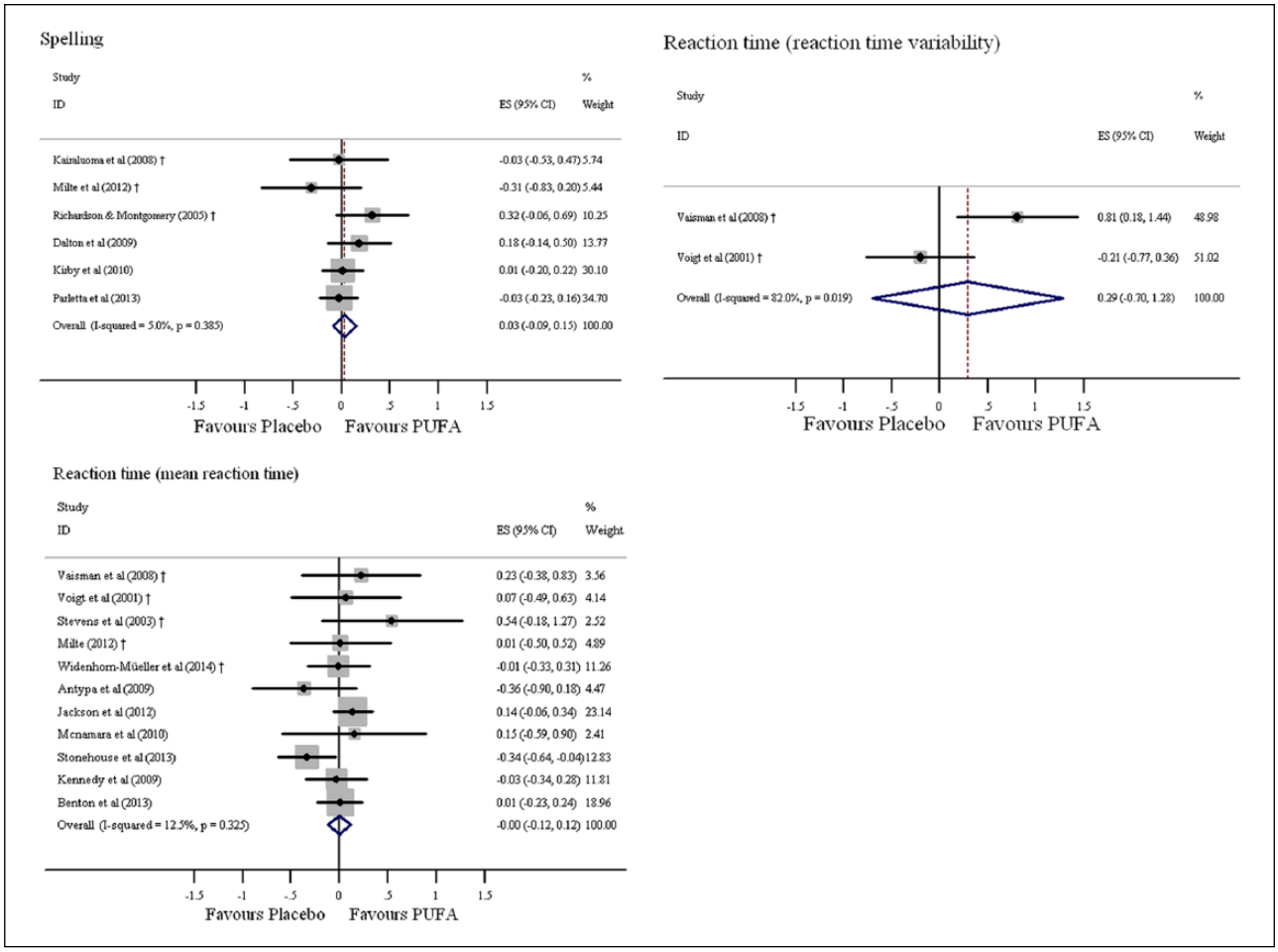


Figure 2. Forest plots for the meta-analyses in the whole sample across the nine domains of cognition. Studies without † indicates typically developing (TD) group. † indicates attention deficit hyperactivity disorder (ADHD)+ related disorder (RD) group.
CI: confidence interval; ES: effect size; ID: identification; IQ: intelligence quotient; PUFA: polyunsaturated fatty acid.

There was no evidence of an effect of *n*-3 PUFA supplementation in TD individuals and those with ADHD+RD. This is in line with the inconsistent findings from individual studies, with few positive findings remaining significant after correction for multiple testing (Antypa et al., 2009; Dalton et al., 2009; Hirayama et al., 2004; Jackson et al., 2012; Parletta et al., 2013; Sinn et al., 2008; Vaisman et al., 2008; Voigt et al., 2001). It is also in line with findings from the three studies included in the qualitative but not quantitative synthesis. Hirayama et al., (2004) found no effect on memory, attention or inhibition after eight weeks of supplementation in children with ADHD. Ryan and Nelson, (2008) found no effect on attention or inhibition after four months supplementation in healthy children. Long and Benton, (2013) found no effect on inhibition after three months supplementation in healthy adult males. This conclusion goes against previous narrative reviews which have suggested *n*-3 PUFA supplementation to improve cognitive performance (Assisi et al., 2006; Bryan et al., 2004; Horrocks and Yeo, 1999; Stonehouse et al., 2013). However, while these reviews highlighted interesting findings, they failed to provide a critical analysis in light of the mixed results on performance measures.

There are several important limitations to be considered before drawing conclusions. This study was limited by substantial between study variation with respect to patient groups, assessment procedures, outcome measures, treatment formulations, and quality in methods adopted for the different studies, necessitating the use of random effects models that produced wider confidence intervals. Due to reporting deficiencies the present study used pre-treatment SD instead of SD of the change (the difference before and after the intervention) in the calculation of effect size (Morris, 2007). This could have resulted in an underestimation of the true effect size (Ortego and Botella, 2010), although a sensitivity analysis of four studies of short-term memory which gave the SD of the change gave a similar, non-significant result (see Supplementary Material, Section S1).

In accord with our predominately negative findings, it has previously been suggested that treatment for ADHD may be more effective for the behavioural symptoms of inattention and hyperactivity-impulsivity, than cognitive performance measures (Coghill et al., 2014). In line with this, previous meta analyses have found a small but significant effect of *n*-3 PUFA supplementation on reducing ADHD symptoms in 699 (SMD=0.31, $p<0.0001$) (Bloch and Qawasmi, 2011) and 827 (SMD=0.21,

p=0.007) (Sonuga-Barke et al., 2013) children with ADHD. Furthermore, meta-analyses and systematic reviews have found a smaller treatment effect of stimulant medication on cognitive performance (~ 0.2–0.6) (Coghill et al., 2014) than on ADHD symptoms (~0.8-1.0) (Banaschewski et al., 2006; Faraone and Buitelaar, 2010). Several recent studies investigating the clinical response to methylphenidate found a dissociation of the treatment effects on ADHD symptoms and cognitive performance in children and adolescents with ADHD (Bédard et al., 2015; Coghill et al., 2007; Schulz et al., 2014). It is therefore suggested that different mechanisms are responsible for change in cognitive performance and change in behavioural symptoms (Coghill et al., 2007).

The lack of significant effects in the ADHD+RD group may reflect neuropsychological heterogeneity leading to a reduced effect size for individual domains of cognitive impairments, in comparison to ADHD symptoms where there is a more uniform deficit (Coghill et al., 2007; Nigg et al., 2005; Sonuga-Barke et al., 2010). For example, Vaisman et al., (2008) included children with a clinical ADHD diagnosis who also performed poorly on a continuous performance test and found a greater number of significant treatment effects on cognitive measures than studies that included those with a clinical ADHD diagnosis regardless of the baseline level of cognitive impairment (for example see Stevens et al., 2003). Although subgroup analyses across four studies which included those with more homogenous cognitive deficits (Kairaluoma et al., 2009; Milte et al., 2012; Richardson et al., 2012; Vaisman et al., 2008) failed to find treatment effects. However, given this small number of studies, further work would be required to test this specific sub-group hypotheses.

The studies used in this meta-analysis varied in supplement composition and dosage according to a previous meta-analysis higher EPA rather than DHA concentrations are associated with symptom reduction in children diagnosed with ADHD (Bloch and Qawasmi, 2011). However a subgroup analysis of those that supplemented with adequate (>100 mg) EPA and a meta-regression examining the relationship between EPA dose and cognitive task performance did little to support this. We found only one small (nominally significant) treatment effect for working memory which was driven by one study (Widenhorn-Müller et al., 2014) and no effect of EPA dose on cognitive performance.

There are inherent problems with blinding in studies which supplement with n-3 PUFA due to the fishy flavour of the capsules. The majority of studies did not assess blinding however above chance guessing occurred in one study that examined this (Milte et al., 2012). Identical flavouring of the placebo and active capsules must be used to reduce this limitation and the possibility of inflated effect sizes. A large number (n=7) of the studies included in the qualitative synthesis used an olive oil placebo. Olive oil contains a high concentration of oleic acid, a precursor of oleamide that has been shown to have psychoactive properties (Richardson, 2006). Stevens et al. (2003) found their olive oil placebo to be 'active' in that the supplement did not maintain the baseline PUFA composition. An inert substance such as liquid paraffin oil could be more suitable (Peet and Horrobin, 2002).

The majority of studies used in this meta-analysis were underpowered. The treatment effect that withstood correction for multiple testing (short-term memory in those who were n-3 PUFA deficient (SMD=0.26)) was small. With this modest effect size of around 0.3 we would require a sample size of around 352 participants (β=80%, two-tailed α=0.05) at a nominal level of significance and around 596 participants after correction for multiple testing (β=80%, two-tailed α=0.006). In the ADHD+RD group trials ranged from 40–362 participants with only three trials above 100. Although the largest trial (Richardson et al., 2012) in healthy children underperforming in reading found treatment effects on reading in only a subgroup of those who were the poorest readers and no effect on working memory. The largest trial in children with ADHD (n=110) (Widenhorn-Müller et al., 2014) again found only marginal evidence of a treatment effect with improvement in working memory but not in six other cognitive performance measures. Future studies should be adequately powered to detect small effects in order to clarify the presence of treatment effects.

We included only school-aged children and adults in our analysis (no trials in adolescent populations were located). The current results are therefore not generalisable to infants, adolescents or the elderly. Research has suggested similar negative results in these groups. A recent meta-analysis examined the effect of n-3 PUFA on cognitive performance in healthy elderly adults and those with cognitive decline. Across 10 domains of cognitive performance, treatment effects were found for those with cognitive decline in three domains (immediate and delayed recall, attention/processing speed). However significance was only at a nominal level (p=0.02–0.04) and became non-significant after correction for multiple testing (Mazereeuw et al., 2012). A Cochrane review and meta-analysis concluded RCTs in infants to have provided little evidence for the effect of n-3 PUFA on neurodevelopmental outcomes (including cognition) and inconsistent effects on visual acuity (Simmer et al., 2011).

Sex dimorphism may also be present in response to PUFA supplementation, thus analysis of samples as a whole and not by sex could potentially mask effects. One study found improvement in episodic memory in women and working memory in men (Stonehouse et al., 2013), potentially reflecting gender differences in problem-solving strategies. However, these findings were not corrected for multiple testing and further evidence would be required to examine the question of sex dimorphism in the cognitive response to n-3 PUFA supplementation.

Length of supplementation has also been proposed as a factor. In the current study only three trials were of six months or longer (Dalton et al., 2009; Osendarp et al., 2007; Stonehouse et al., 2013). Across two of these studies treatment effects were found on verbal learning ability, memory and reaction time (Dalton et al., 2009; Stonehouse et al., 2013). Although Stonehouse et al. (2013) tested a large number of cognitive domains, the majority of which were non-significant and failed to correct for multiple testing. The longest study (12 months) failed to find any treatment effects (albeit the dosage of n-3 PUFA was relatively small; Osendarp et al., 2007). The current study found no relationship between length of supplementation and effects on cognitive performance which is in line with a recent meta-analysis that found no relationship between trial duration and efficacy of n-3 PUFA supplementation in reducing ADHD symptoms (Bloch and Qawasmi, 2011). This evidence suggests that outcomes may have been uninfluenced by duration.

A number of outcome measures such as accuracy on cognitive tasks could not be included in this analysis due to the measures being too diverse to combine. However results from such measures were largely negative. For example treatment effects were not found in TD children (Kennedy et al., 2009) and adults (Jackson et al., 2012) for accuracy on reaction time tasks or in

children with ADHD for speed of information processing tasks (Widenhorn-Müller et al., 2014). Although one study (Sinn et al., 2008) found a significant benefit of treatment for accuracy on a sustained attention task, overall these results are in line with current negative findings.

In conclusion we have found no evidence of an effect of *n*-3 PUFA supplementation on cognitive performance in the general population or in those with ADHD and related disorders. There was suggestive evidence of improvements in those with low *n*-3 PUFA status. In order to provide a more conclusive picture future trials should employ larger sample sizes and should focus on supplementation of those who are *n*-3 PUFA deficient. It is suggested that regulators and producers of omega-3 products should consider this evidence when promoting their products.

Acknowledgements

The authors would like to thank Charlotte Scott for her assistance with data extraction, Rob Power for proof reading the manuscript and Vifor Pharma who provided the grant used to support REC. They are grateful to the authors of the included studies who took the time to provide us with the requested data.

Declaration of Conflicting Interests

The authors declared the following potential conflicts of interest with respect to the research, authorship, and/or publication of this article: Ruth Cooper is a PhD student funded by Vifor Pharma. Philip Asherson received funds for consultancy or sponsored talks on behalf of Kings College London (KCL) for Shire, Lilly, Novartis, Janssen and PCM Scientific. He received research or education funds on behalf of KCL from Shire, Lilly, Novartis, Janssen, Vifor Pharma and QbTech. Charlotte Tye, Jonna Kuntsi and Evangelos Vassos declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding

The authors disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: This work was supported by Vifor Pharma (grant number: PADWUDB) who provided funds for a student grant to PA, used to support REC. The work was completed by the ADHD research group at the MRC Social Genetic and Developmental Psychiatry (SGDP) Centre, Kings College London. Vifor Pharma, played no role in the conceptualisation, analysis, interpretation or writing of the meta-analysis. REC had full access to all the study data and had final responsibility for the decision to submit for publication.

References

Aberg MAL, Aberg N, Brisman J, et al. (2009) Fish intake of Swedish male adolescents is a predictor of cognitive performance. *Acta Paediatr* 98: 555–560.

Amminger GP, Schafer MR, Papageorgiou K, et al. (2010) Long-chain -3 fatty acids for indicated prevention of psychotic disorders. *Arch General Psychiatry* 67: 146–154.

Antypa N, Van der Does AJW, Smelt AHM, et al. (2009) Omega-3 fatty acids (fish-oil) and depression-related cognition in healthy volunteers. *J Psychopharmacol* 23: 831–840.

Assisi A, Banzi R, Buonocore C, et al. (2006) Fish oil and mental health: The role of n-3 long-chain polyunsaturated fatty acids in cognitive development and neurological disorders. *Int Clin Psychopharmacol* 21: 319–336.

Banaschewski T, Coghill D, Santosh P, et al. (2006) Long-acting medications for the hyperkinetic disorders. A systematic review and European treatment guideline. *Eur Child Adolesc Psychiatry* 15: 476–495.

Baumgartner J, Smuts CM, Malan L, et al. (2012) Effects of iron and n-3 fatty acid supplementation, alone and in combination, on cognition in school children: A randomized, double-blind, placebo-controlled intervention in South Africa. *Am J Clin Nutr* 96: 1327–1338.

Bédard A-CV, Stein MA, Halperin JM, et al. (2015) Differential impact of methylphenidate and atomoxetine on sustained attention in youth with attention-deficit/hyperactivity disorder. *J Child Psychol Psychiatry* 56: 40–48.

Benton D, Donohoe RT, Clayton DE, et al. (2013) Supplementation with DHA and the psychological functioning of young adults. *Br J Nutr* 109: 155–161.

Bloch MH and Qawasmi A (2011) Omega-3 Fatty acid supplementation for the treatment of children with attention-deficit/hyperactivity disorder symptomatology: Systematic review and meta-analysis. *J Am Acad Child Adolesc Psychiatry* 50: 991–1000,

Bryan J, Ph D, Osendarp S, et al. (2004) Nutrients for cognitive development in school-aged children. *Nutr Rev* 62: 295–306.

Coghill DR, Rhodes SM and Matthews K (2007) The neuropsychological effects of chronic methylphenidate on drug-naive boys with attention-deficit/hyperactivity disorder. *Biol Psychiatry* 62: 954–962.

Coghill DR, Seth S, Pedroso S, et al. (2014) Effects of methylphenidate on cognitive functions in children and adolescents with attention-deficit/hyperactivity disorder: Evidence from a systematic review and a meta-analysis. *Biol Psychiatry* Elsevier, 76: 603–615.

Cohen J (1988) *Statistical Power Analysis for the Behavioural Sciences*. 2nd ed. Hillsdale New Jersey: Erlbaum.

Dalton A, Wolmarans P, Witthuhn RC, et al. (2009) A randomised control trial in schoolchildren showed improvement in cognitive function after consuming a bread spread, containing fish flour from a marine source. *Prostaglandins Leukot Essent Fatty Acid* 80: 143–149.

Doyle AE, Willcutt EG, Seidman LJ, et al. (2005) Attention-deficit/hyperactivity disorder endophenotypes. *Biol Psychiatry* 57: 1324–1335.

Dunnick JK and Hailey JR (1995) Experimental studies on the long-term effects of methylphenidate hydrochloride. *Toxicology* 103: 77–84.

Elbourne DR, Altman DG, Higgins JPT, et al. (2002) Meta-analyses involving cross-over trials: Methodological issues. *Int J Epidemiol* 31: 140–149.

Faraone S V and Buitelaar J (2010) Comparing the efficacy of stimulants for ADHD in children and adolescents using meta-analysis. *Eurn Child Adolesc Psychiatry* 19: 353–364.

Field AP and Gillett R (2010) How to do a meta-analysis. *Br J Math Stat Psychol* 63: 665–694.

Frazier TW, Demaree HA and Youngstrom EA (2004) Meta-analysis of intellectual and neuropsychological test performance in attention-deficit/hyperactivity disorder. *Neuropsychology* 18: 543–555.

Goyens PLL, Spilker ME, Zock PL, et al. (2005) Compartmental modeling to quantify alpha-linolenic acid conversion after longer term intake of multiple tracer boluses. *J Lipid Res* 46: 1474–1483.

Greenberg LM and Kindschi CL (1996) *T.O.V.A. Test of Variables of Attention: Clinical Guide*. Los Alamitos, California: Universal Attention Disorders Inc.

Gustafsson P, Birberg-Thornberg U, Duchén K, et al. (2010) EPA supplementation improves teacher-rated behaviour and oppositional symptoms in children with ADHD. *Acta Paediatr* 99: 1540–1549.

Hamazaki T, Sawazaki S, Itomura M, et al. (1996) Rapid publication the effect of docosahexaenoic acid on aggression in young adults. *J Clin Invest* 97: 1129–1133.

Hibbeln JR, Davis JM, Steer C, et al. (2007) Maternal seafood consumption in pregnancy and neurodevelopmental outcomes in childhood (ALSPAC study): An observational cohort study. *Lancet*,369: 578–585.

Higgins JPT and Green S (eds) (2011) *Cochrane Handbook for Systematic Reviews of Interventions* Version 5.1.0 [updated March

2011]. The Cochrane Collaboration. Available at www.cochrane-handbook.org

Hirayama S, Hamazaki T and Terasawa K (2004) Effect of docosahexaenoic acid-containing food administration on symptoms of attention-deficit/hyperactivity disorder – a placebo-controlled double-blind study. *Eur J Clin Nutr* 58: 467–473.

Horrocks L a and Yeo YK (1999) Health benefits of docosahexaenoic acid (DHA). *Pharmacoll Res* 40: 211–225.

Jackson P, Deary ME, Reay JL, et al. (2012) No effect of 12 weeks' supplementation with 1 g DHA-rich or EPA-rich fish oil on cognitive function or mood in healthy young adults aged 18-35 years. *Br J Nutr* 107: 1232–1243.

Kairaluoma L, Närhi V, Ahonen T, et al. (2009) Do fatty acids help in overcoming reading difficulties? A double-blind, placebo-controlled study of the effects of eicosapentaenoic acid and carnosine supplementation on children with dyslexia. *Child Care Health Dev* 35: 112–119.

Kalmijn S, van Boxtel MPJ, Ocké M, et al. (2004) Dietary intake of fatty acids and fish in relation to cognitive performance at middle age. *Neurology* 62: 275–280.

Karr JE, Grindstaff TR and Alexander JE (2012) Omega-3 polyunsaturated fatty acids and cognition in a college-aged population. *Exp Clin Psychopharmacol* 20: 236–242.

Kennedy DO, Jackson P a, Elliott JM, et al. (2009) Cognitive and mood effects of 8 weeks' supplementation with 400 mg or 1000 mg of the omega-3 essential fatty acid docosahexaenoic acid (DHA) in healthy children aged 10-12 years. *Nutr Neurosci* 12: 48–56.

Kirby A, Woodward A, Jackson S, et al. (2010) A double-blind, placebo-controlled study investigating the effects of omega-3 supplementation in children aged 8-10 years from a mainstream school population. *Res Dev Disabil* 31: 718–730.

Kuntsi J, Wood AC, Van Der Meere J, et al. (2009) Why cognitive performance in ADHD may not reveal true potential: Findings from a large population-based sample. *J Int Neuropsychol Soc JINS* 15: 570–579.

Leonard BE, McCartan D, White J, et al. (2004) Methylphenidate: A review of its neuropharmacological, neuropsychological and adverse clinical effects. *Hum Psychopharmacol* 19: 151–180.

Long S and Benton D (2013) A double-blind trial of the effect of docosahexaenoic acid and vitamin and mineral supplementation on aggression, impulsivity, and stress. *Hum Psychopharmacol* 28: 238–247.

Mazereeuw G, Lanctôt KL, Chau S a, et al. (2012) Effects of ω-3 fatty acids on cognitive performance: A meta-analysis. *Neurobiol Aging* 33: 1482.e17–29.

McNamara RK, Able J, Jandacek R, et al. (2010) Docosahexaenoic acid supplementation increases prefrontal cortex activation during sustained attention in healthy boys: A placebo-controlled, dose-ranging, functional magnetic resonance imaging study. *Am J Clin Nutr* 91: 1060–1067.

Milte CM, Parletta N, Buckley JD, et al. (2012) Eicosapentaenoic and docosahexaenoic acids, cognition, and behavior in children with attention-deficit/hyperactivity disorder: A randomized controlled trial. *Nutrition* 28: 670–677.

Moher D, Liberati A, Tetzlaff J, et al. (2009) Preferred reporting items for systematic reviews and meta-analyses: The PRISMA statement. *PLoS medicine*, 6: e1000097.

Morris SB (2007) Estimating effect sizes from pretest-posttest-control group designs. *Organizational Research Methods*, 11: 364–386.

Nasrallah H A, Loney J, Olson SC, et al. (1986) Cortical atrophy in young adults with a history of hyperactivity in childhood. *Psychiatry Res* 17: 241–246.

Nigg JT, Willcutt EG, Doyle AE, et al. (2005) Causal heterogeneity in attention-deficit/hyperactivity disorder: Do we need neuropsychologically impaired subtypes? *Biol Psychiatry* 57: 1224–1230.

Ortego C and Botella J (2010) The hard but necessary task of gathering order-one effect size indices in meta-analysis. *Psicológica* 31: 291–315.

Osendarp SJM, Baghurst KI, Bryan J, et al. (2007) Effect of a 12-mo micronutrient intervention on learning and memory in well-nourished and marginally nourished school-aged children: 2 Parallel, randomized, placebo-controlled studies in Australia and Indonesia. *Am J Clin Nutr* 86: 1082–1093.

Parletta N, Cooper P, Gent DN, et al. (2013) Effects of fish oil supplementation on learning and behaviour of children from Australian Indigenous remote community schools: A randomised controlled trial. *Prostaglandins Leukot Essent Fatty Acid* 89: 71–79.

Peet M and Horrobin DF (2002) A dose-ranging study of the effects of ethyl-eicosapentaenoate in patients with ongoing depression despite apparently adequate treatment with standard drugs. *Arch Gen Psychiatry* 59: 913–919.

Portillo-Reyes V, Pérez-García M, Loya-Méndez Y, et al. (2014) Clinical significance of neuropsychological improvement after supplementation with omega-3 in 8-12 years old malnourished Mexican children: A randomized, double-blind, placebo and treatment clinical trial. *Res Dev Disabil* 35: 861–870.

Richardson AJ (2006) Omega-3 fatty acids in ADHD and related neurodevelopmental disorders. *Int Rev Psychiatry* 18: 155–172.

Richardson AJ and Montgomery P (2005) The Oxford-Durham study: A randomized, controlled trial of dietary supplementation with fatty acids in children with developmental coordination disorder. *Pediatrics* 115: 1360–1366.

Richardson AJ, Burton JR, Sewell RP, et al. (2012) Docosahexaenoic acid for reading, cognition and behavior in children aged 7-9 years: A randomized, controlled trial (the DOLAB Study). *PloS One* 7: e43909.

Ryan AS and Nelson EB (2008) Assessing the effect of docosahexaenoic acid on cognitive functions in healthy, preschool children: A randomized, placebo-controlled, double-blind study. *Clin Pediatr (Phila)* 47: 355–362.

Schulz KP, Fan J and Be A V (2014) Common and unique therapeutic mechanisms of stimulant and nonstimulant treatments for attention-deficit/hyperactivity disorder. *Arch Gen Psychiatry* 69: 952–961.

Simmer K, Patole SK and Rao SC (2011) Longchain polyunsaturated fatty acid supplementation in infants born at term. *Cochrane Database Syst Rev* 12: 1–72.

Sinn N, Bryan J and Wilson C (2008) Cognitive effects of polyunsaturated fatty acids in children with attention deficit hyperactivity disorder symptoms: A randomised controlled trial. *Prostaglandins Leukot Essent Fatty Acid* 78: 311–326.

Sonuga-Barke E, Bitsakou P and Thompson M (2010) Beyond the dual pathway model: Evidence for the dissociation of timing, inhibitory, and delay-related impairments in attention-deficit/hyperactivity disorder. *J Am Acad Child Adolesc Psychiatry* 49: 345–355.

Sonuga-Barke EJS, Brandeis D, Cortese S, et al. (2013) Nonpharmacological interventions for ADHD: Systematic review and meta-analyses of randomized controlled trials of dietary and psychological treatments. *Am J Psychiatry* 170: 275–289.

StataCorp (2009) *Stata Statistical Software: Release 11*. College Station, Texas: StataCorp LP.

Stevens L, Zhang W, Peck L, et al. (2003) EFA supplementation in children with inattention, hyperactivity, and other disruptive behaviors. *Lipids* 38: 1007–1021.

Stonehouse W, Conlon CA, Podd J, et al. (2013) DHA supplementation improved both memory and reaction time in healthy young adults: A randomized controlled trial. *Am J Clin Nutr* 97: 1134–1143.

Su K-P, Lai H-C, Yang H-T, et al. (2014) Omega-3 fatty acids in the prevention of interferon-alpha-induced depression: Results from a randomized, controlled trial. *Biol Psychiatry* 76: 559–566.

Vaisman N, Kaysar N, Zaruk-adasha Y, et al. (2008) Correlation between changes in blood fatty acid composition and visual sustained attention performance in children with inattention. *Am J Clin Nutr* 87: 1170–1180.

Voigt RG, Llorente AM, Jensen CL, et al. (2001) A randomized, double-blind, placebo-controlled trial of docosahexaenoic acid

supplementation in children with attention-deficit/hyperactivity disorder. *Journal of Pediatrics* 139: 189–196.

Wechsler D (1991) *Manual for the Wechsler Scale of Children's Intelligence-III*. New York: Psychological Corporation.

Widenhorn-Müller K, Schwanda S, Scholz E, et al. (2014) Effect of supplementation with long-chain ω-3 polyunsaturated fatty acids on behavior and cognition in children with attention deficit/hyperactivity disorder (ADHD): A randomized placebo-controlled intervention trial. *Prostaglandins Leukot Essent Fatty Acids* 91: 49–60.

Willcutt EG, Pennington BF, Duncan L, et al. (2010) Understanding the complex etiologies of developmental disorders: Behavioral and molecular genetic approaches. *J Dev Behav Pediatr* 31: 533–544.

Wilkinson G and Roberts G (2006) *Wide Range Achievement Test, Fourth Edition*. Florida: Psychological Assessment Resources.

Yui K, Koshiba M, Nakamura S, et al. (2012) Effects of large doses of arachidonic acid added to docosahexaenoic acid on social impairment in individuals with autism spectrum disorders: A double-blind, placebo-controlled, randomized trial. *J Clin Psychopharmacol* 32: 200–206.