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

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Abstract: Autism spectrum disorders are a neurodevelopmental disorders with reduced cortical functional connectivity relating to social cognition. Polyunsaturated fatty acids arachidonic acid (ARA) and docosahexaenoic acid (DHA) may have key role in brain network maturation. In particularly, ARA is important in signal transduction related to neuronal maturation. Supplementation with larger ARA doses added to DHA may therefore mitigate social impairment. In a 16-week, double-blind, randomized, placebo-controlled trial, we evaluated the efficacy of supplementation with large doses of ARA added to DHA (n = 7) or placebo (n = 6) in 13 participants (mean age, 14.6 [SD, 5.9] years). To examine underlying mechanisms underlying the effect of our supplementation regimen, we examined plasma levels of antioxidants transferrin and superoxide dismutase, which are useful markers of signal transduction. The outcome measures were the Social Responsiveness Scale and the Aberrant Behavior Checklist–Community. Repeated-measures analysis of variance revealed that our supplementation regimen significantly improved Aberrant Behavior Checklist–Community–measured social withdrawal and Social Responsiveness Scale–measured communication. Treatment effect sizes were more favorable for the treatment group compared with the placebo group (communication: treatment groups, 0.87 vs, placebo, 0.44; social withdrawal: treatment groups, 0.88, vs placebo, 0.54). There was a significant difference in the change in plasma transferrin levels and a trend toward a significant difference in the change in plasma superoxide dismutase levels between the 2 groups. This preliminary study suggests that supplementation with larger ARA doses added to DHA improves impaired social interaction in individuals with autism spectrum disorder by up-regulating signal transduction.

Key Words: autism spectrum disorders, dietary unsaturated fatty acids, arachidonic acid, docosahexaenoic acid, social interaction

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Autism spectrum disorders (ASDs) are characterized by severe deficits in socialization, communication, and disruptive repetitive behaviors. Previous double-blind, randomized, placebo-controlled studies have reported that atypical antipsychotic agent risperidone[1–3] or selective serotonin reuptake inhibitors (eg, fluoxetine and fluvoxamine)[4] significantly reduced disruptive repetitive behaviors. They do not provide any clear benefit with regarding to impaired social interaction.[1,2] In a previous study, a brief period of risperidone treatment gave rise to adverse events (such as weight gain and tremor), but the short period of this trial limits inferences about tardive dyskinesia.[1] There is no significant difference in frequency of the extrapyrmidal symptoms between risperidone and placebo.[5] Selective serotonin reuptake inhibitor occasionally induced extrapyrmidal symptoms.[6] It is important to consider possible adverse effects of these drugs, in particular weight gain, fatigue, agitation, irritability,[3,4] and tardive dyskinesia.[1] Treatments are needed for the core social impairments of ASD that do not have adverse effects.

Magnetic resonance imaging studies have suggested that reduced long-distance functional connectivity, in the networks that underlie socioemotional and communication functions, is related to the social impairment that is observed in ASD.[7] This reduced long-distance connectivity might be due to inadequate numbers and timing of long-distance afferents, which would affect the delivery of afferent signals to higher-order cortical regions.[7] The central nervous system is rich in highly polyunsaturated fatty acids (PUFAs), particularly docosahexaenoic acid (DHA), which is an omega-3 fatty acid, and arachidonic acid (ARA), an omega-6 fatty acid. Docosahexaenoic acid and ARA are essential for neurodevelopment.[8,9] Links have been reported between ASD and omega-6–omega-3 fatty acid imbalances or impaired PUFA metabolism.[10–12] Most studies of dietary PUFAs supplementation have therefore examined combinations of high doses of omega-3 fatty acids (eicosapentaenoic acid [EPA], DHA) and omega-6 fatty acids (ARA). Although supplementation with DHA alone[13] or with EPA plus DHA[14] did not produce significant clinical improvements in ASD, these open-label trials provide limited interference about negative effects. Previous double-blind, randomized, placebo-controlled studies reported that PUFA supplementation improved behavioral problems in children with attention-deficient/hyperactivity disorder (ADHD),[9] with learning difficulties,[15] and with ASD.[16] Arachidonic acid is important in a number of brain processes, including signal transduction.[17,18] Infant formula with 0.36% DHA and 0.72% ARA promotes mental development,[19] and ARA-containing diets assist in neurogenesis in the hippocampus of mice.[20] Arachidonic acid may therefore mitigate social impairment in individuals with ASD. Unfortunately, there is a paucity of randomized controlled studies investigating the effects in ASD of large doses of ARA added to DHA on social impairment. Even though behavioral intervention is the usual form of treatment, we preferred

to conduct a double-blind, randomized, placebo-controlled trial to measure the effects on social impairment in ASD of larger doses of ARA added to DHA supplementation. To compensate for our small sample size, we used a life-span approach by including participants with a large age range.

Consideration of a plausible mechanism of action may be helpful for estimating the efficacy of most of complementary and alternative medicine therapies.[21] It is well known that omega-3 fatty acids[22] and ARA[17] have a direct action on signal transduction, including neurotransmission. There is increasing evidence that the lipid antioxidant transferrin (TF) is important in signal transduction related to neuronal development[23,24]; also, and superoxide dismutase (SOD) acts as a modifier of redox signaling related to oxidative stress during neuronal development.[25,26] Superoxide dismutase and TF may therefore be useful markers of signal transduction. In the present study, we examined the plasma levels of SOD, TF, and PUFAs such as EPA, DHA, and ARA to clarify the mechanism responsible for the effects of our supplementation regimen.

MATERIALS AND METHODS

Patients

The study population was composed of 13 individuals (12 males and 1 female) in the age range 6 to 28 years (mean age, 14.6 [SD, 6.0] years) who had autistic disorder, showing delays in the development of spoken language (n = 1) or Asperger disorder (n = 12), according to the patient version of the Structured Clinical Interview for *Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition*[27]; diagnosis was corroborated using a standardized semistructured interview, the Autism Diagnostic Interview–Revised.[28] Each diagnosis was established at screening by the agreement of 2 independent psychiatrists. Of the 13 participants, 4 were aged 6 to 10 years, 8 were aged 13 to 20 years, and 1 participant was 28 years old. They were assigned randomly to receive either the experimental medication (n = 7; mean age, 13.9 [SD, 5.3] years; range, 6–20 years) or a placebo (n = 6; mean age, 15.5 [SD, 7.4] years; range, 7-18 years, plus a 28-year-old). The participants were recruited to Ashiya University's Research Institute of Pervasive Developmental Disorders between October 2008 and January 2009 via a local advertisement. A physical examination (sitting blood pressure, heart rate, weight, and height) and clinical laboratory measurements (clinical chemistry, platelet activation, hematology, and urine toxicology) were conducted by 2 psychiatrists, at screening and at the end of the trial. The criteria for inclusion were as follows: (1) free of medical or comorbid psychiatric disorders, (2) weight of at least 16 kg, and (3) verbal or performance IQ greater than 80 at baseline as measured by the Wechsler Intelligence Scale for children and adolescents aged 6 to 16 years[29] or for adults[30] (our third criterion is because total IQ is at least 80 in subjects with Asperger disorders[31]), and (4) a score of greater than 10 on the social withdrawal subscale of the Aberrant Behavior Checklist (ABC)[32] at baseline. The fourth criterion is because the mean score on the social withdrawal subscale in subjects aged 5 to 30 years was found to be less than 10 in 531 subjects with neuropsychiatric diseases (eg, deafness and psychoses).[33] Individuals who were expected to require hospitalization for their behavioral symptoms were excluded, as were those with clinically significant abnormal laboratory data or who had been treated with antidepressants, anxiolytic medication, or neuroleptics within the past 6 months. Treatment of ADHD symptoms with stimulants was allowed during the study provided that the patient's dosage had remained stable for at least 3 months before and during the study. Any participant who received a program teaching appropriate social skills within the previous 3 months was excluded.

The study was conducted at the Department of Child and Adolescent Psychiatry, Sawa Hospital, which is a psychiatric institute of Ashiya University in Osaka. The ethics committee of Sawa Hospital approved the study protocol. The committee was given a full description of the study, after which written informed consent was obtained from the participant's parents, the participant, or both. To verify adherence to protocol, clinical monitors examined the source documents, and all collected were reviewed for protocol deviations.

Supplementary Material

Commercially available Aravita (Suntory Ltd, Osaka, Japan) contains 40 mg/capsule of DHA, 40 mg/capsule of ARA, and 0.16 mg/capsule of astaxanthin (a PUFA antioxidant). We purchased Aravita and identical capsules containing olive oil from Suntory Ltd (Supplemental Chart, Supplemental Digital Content 1, which shows Consort Flowchart, http://links.lww.com/JCP/A114).

Study Design

After screening, the 13 eligible participants were assigned to either a supplementation with larger doses of ARA added to DHA (n = 7) or a matching placebo (n = 6) according to a block-balanced randomized plan based on their demographic data, medical and psychiatric history, routine laboratory evaluation results, and also the baseline scores for the 2 outcome measures, that is, the Social Responsiveness Scale (SRS)[34] and the ABC.[32] A random allocation sequence (1:1) was generated by 2 pharmacological scientists who had no direct relation to the study. As a result, the investigators, subjects, and research staff were blind to which group each subject had been assigned throughout the study.

In a double-blind, randomized, placebo-controlled trial conducted for 16 weeks, the daily experimental medication was 240 mg of ARA-enriched triglyceride (SUNTGA40S, containing 40 mg/capsule each of ARA and DHA, and 0.16 mg/capsule of astaxanthin in capsule measuring 6 × 9 mm). The usual daily doses of this capsule were 6 capsules (240 mg). The placebo was an identical capsule containing olive oil. Four participants aged 6 to 10 years received a daily supplementation dose of 120 mg.

Efficacy Assessment

Clinical outcome evaluations were carried out at baseline and at 4, 8, 12, and 16 weeks after the intervention, using the SRS[34] and the ABC.[32] The SRS and ABC subscales were completed by the parent. The SRS is a 65-item quantitative measure of autistic social impairment[35] completed by an informant. An adult version of the SRS exists for subjects 19 years or older and was used in this study. The ABC was intended primarily to evaluate treatment in ASD psychopharmacological and behavioral intervention trials involving children and adolescents with normal IQ levels.[36] It consists of 58 items, each of which is scored on a 4-point scale. The items fall into 5 subscales: (1) irritability, (2) social withdrawal, (3) stereotypy, (4) hyperactivity, and (5) inappropriate speech.[32]

Safety Assessment

Throughout the study, safety of the supplementation regimen was assessed by monitoring and recording at the time each event was reported. Adverse events were either spontaneously reported by the participants or the participants' guardians or

were noted by the investigator. Questioning focused on known adverse effects of ARA, followed by open-ended questioning. Adverse effects that were investigated included nausea, vomiting, stomach ache, and diarrhea.

Assays of Plasma Levels of PUFAs, SOD, and TF

Plasma samples were obtained from blood anticoagulated with EDTA and frozen at −80°C until analysis. All assays were carried out by specialists at SRL, Inc, Tokyo, Japan.

Plasma Levels of PUFAs

The fatty acid composition of the total phospholipid fraction of the patients' plasma was determined as follows.[37] In summary, total lipids were extracted from the plasma according to the method of Bligh and Dyer.[38] After transmethylation with HCl-methanol, the fatty acid composition was analyzed by gas chromatography (GC2010; Shimadzu Co, Tokyo, Japan). A total of 24 long-chain fatty acids were identified. The sensitivity of detection of our method for measuring the plasma levels of DHA and ARA was 0.2 μg/mL. The intra-assay and interassay coefficients of ARA were 110.14 μg/mL (SD, 3.87; CV, 5.28%) and 100.63 μg/mL (SD, 5.51; CV, 5.48%), respectively, and those of DHA were 73.87 μg/mL (SD, 2.30; CV, 3.11%) and 68.07 μg/mL (SD, 2.30; CV, 3.33%), respectively. The plasma level of each PUFA is expressed as the percentage weight of total fatty acids as mean (SD).

Transferrin

Plasma TF levels were determined using a standard turbidimetric assay and an automated biochemical analyzer (JCA-BM8000 series; JEOL Ltd, Tokyo, Japan). The minimum detectable concentration was 21.0 mg/dL. The intra-assay and interassay coefficients were 108.1 and 107.4 mg/dL, respectively.

Superoxide Dismutase

Plasma SOD levels were estimated from the rate of decrease in nitrite produced by hydroxylamine and superoxide anions, based on nitrite method[39] by using VERSA max (Molecular Devices Co, Tokyo, Japan). The human plasma was assayed using the SOD assay kit according to the cytochrome *c* method. Plasma SOD levels are expressed as units per milliliter. The assay sensitivity was 0.3 U/mL. The intra-assay and interassay coefficients were 2.11 and 2.10 U/mL, respectively.

Statistical Analyses

Sample size was calculated with G*Power 3.1.2 (SPSS Marketing Software) for the within-between interaction in analysis of variance (ANOVA) (input parameter: 0.70 effect size with 80% power, and a 0.05 significance level in a 2-sided test). Repeated-measures ANOVA is usually used to eliminate or reduce individual differences as a source of between-group differences, and it is the most appropriate statistical test for the present study.[40] To compare the ABC and SRS scores between the 2 groups, repeated-measures ANOVA was undertaken with baseline scores as the covariate, treatment as the between-subject factor, and test session as a repeated factor, so as to adjust for pretreatment differences. The data were not normally distributed, so that the group comparisons at baseline and at 16 weeks were carried out using the Mann-Whitney *U* test. The χ^2 test was used for categorical variables. All tests of hypotheses were performed using a 2-sided significance levels of 0.05, whereas *P* between 0.050 and 0.10 is taken as a trend toward significance.[41] To analyze the data, we used SPSS version 18. Adverse events were recorded with the *Medical Dictionary for Regulatory Activities* (version 13.1).[42]

RESULTS

Subject Characteristics

Total scores of SRS and ABC at baseline in the 13 participants were 138.1 (SD, 29.1) and 74.5 (SD, 30.4), respectively. The total SRS and ABC score in children and adolescents with ASD were 101.7 (SD, 22.1),[43] and the corresponding ABC score was 85.6 (SD, 27.3),[44] and because total SRS scores greater than 80 indicate mild to moderate interference in social interaction,[35] our patients were all considered to have severe conditions.

The participants were assigned randomly to receive either the experimental medication (n = 7; aged 6, 8, 13, 14, 17, 19, and 20 years; mean age, 13.9 [SD, 5.3] years) or placebo (aged 7, 10, 13, 17, 18, and 28 years; mean age, 15.5 [SD, 7.4] years). There were no significant differences between the group in age ($U = 20.0$, $P = 0.95$) or in baseline scores of the SRS and ABC subscale scores; all $P > 0.05$ (Table 1).

Efficacy Results

In the treatment group, ABC social withdrawal scores ($F = 3.08$, $P < 0.01$) and SRS communication subscale scores ($F = 5.23$, $P < 0.05$) improved significantly over the 16-week supplementation period relative to the placebo group (Tables 2 and 3). At the end of the study, the ABC social withdrawal scores of the treatment group were significantly improved better than those of the placebo group ($U = 6.0$, $P < 0.05$). The numbers of individuals who achieved 50% improvements in the total SRS (treatment group: 14.3% vs placebo group: 16.7%, $\chi^2 = 0.01$, $P = 0.92$) or ABC (treatment group: 42.9% vs placebo group: 16.7%, $\chi^2 = 0.57$, $P = 0.45$) score from the baseline did not differ significantly between the 2 groups, but a greater portion of people in the treatment group exhibited a 50% improvement in their ABC total scores than the in the placebo group. The treatment effect sizes for ABC social withdrawal (treatment group: 0.88 vs placebo: 0.54) and SRS communication (treatment group: 0.87 vs placebo: 0.44) subscales were more favorable for the treatment group than for the placebo group.

Adverse Events

No adverse events were reported in either group.

TABLE 1. Demographic Comparison Between the Treatment Group and the Placebo Group at the Baseline

	Group			
	Supplementation (n = 7)	Placebo (n = 6)	*U*	*P**
Age, y (mean±SD)	10.1 (3.2)	19.8 (4.2)	20.0	0.95
SRS score (mean±SD)				
Awareness	14.6 (2.8)	17.3 (5.2)	11.0	0.18
Cognition	24.9 (5.8)	25.2 (6.2)	21.0	1.00
Communication	46.9 (12.6)	51.3 (8.8)	17.0	0.63
Motivation	22.7 (9.9)	25.3 (5.0)	14.0	0.37
Mannerisms	23.9 (9.4)	22.5 (7.3)	17.0	0.63
ABC score (mean±SD)				
Irritability	12.8 (10.8)	12.7 (7.6)	16.5	0.53
Social withdrawal	26.7 (10.7)	30.0 (7.9)	16.0	0.53
Stereotypy	7.6 (7.8)	8.0 (6.8)	21.0	1.00
Hyperactivity	18.3 (9.9)	20.3 (15.5)	19.5	0.84
Inappropriate speech	7.3 (6.8)	4.5 (4.9)	18.0	0.73

*Mann-Whitney *U* test.

TABLE 2. Mean Changes (±SD) From Baseline to End Point Scores

	Group		Statistical Differences			
Entire Group	**Supplementation (n = 7)**	**Placebo (n = 6)**	***U***	***P****	**χ^2**	***P*†**
SRS total score						
Baseline	133.0 (31.2)	140.0 (27.4)	13.0	0.30		
End point	89.3 (28.4)	93.2 (13.2)	20.5	0.95		
No. with 50% improvement	1/7	1/6			0.57	0.45
ABC total score						
Baseline	72.1 (27.5)	75.0 (38.8)	21.0	1.00		
End point	38.3 (28.2)	52.7 (22.5)	9.0	0.10		
No. with 50% improvement	3/7	1/6			0.01	0.92

*Mann-Whitney *U* test.

†χ^2 Analysis.

Plasma Levels of PUFAs, SOD, and TF

Repeated-measures ANOVA, controlling for baseline severity, found a significant difference ($F = 7.362$, $P < 0.05$) in the change in plasma TF levels between the 2 groups. There was also a trend toward significant difference ($F = 3.77$, $P = 0.08$) in the change in plasma SOD levels. There were no significant differences in the plasma levels of DHA ($F = 0.12$, $P = 0.74$) or ARA ($F = 0.033$, $P = 0.86$). At the end of the study there were no significant differences in the plasma levels of PUFAs, SOD, or TF (Table 4).

DISCUSSION

These are the first findings of a double-blind, randomized, placebo-controlled trial that examines whether adding larger doses of ARA to DHA supplementation improved social withdrawal and communication symptoms in individuals with ASD. Previous placebo-controlled trials of supplementation with DHA, EPA, and ARA yielded significant improvement in 7 of 14 scales of behavioral problems among children with learning difficulties[9] and 2 of 16 scales of behavioral problems among children with ADHD.[15] In another placebo-controlled trial, higher doses of EPA plus DHA supplementation produced a trend toward a significant improvement in the ABC hyperactivity symptoms of the 5 ABC subscales in 12 children with ASD.[16] The effect found in the present study are small compared with previous studies,[9,15,16] but we find improvement in social interaction. Docosahexaenoic acid and ARA play key roles in neuronal maturation.[8,18] Omega-6–omega-3 fatty acid imbalances or alterations in PUFA composition have been found in ASD.[10–12] The dose ratio of ARA to DHA in the supplementation trial may therefore be important. The ARA/DHA ratios in the 2 previous clinical studies were 0.088 (Richardson and Puri[9]) or 0.083 (Stevens et al[15]). On the other hand, ARA/DHA ratio in this study

TABLE 3. Mean Scores (±SD) on the ABC and the SRS for the 16-Week Trial

	Groups									
	Supplementation (n = 7)				Placebo (n = 6)					
Outcome Measures	**4 wk**	**8 wk**	**12 wk**	**16 wk**	**4 wk**	**8 wk**	**12 wk**	**16 wk**	**F**	***P****
SRS score										
Awareness	10.9 (2.7)	10.3 (2.9)	13.0 (2.2)	7.3 (3.6)	9.8 (2.6)	10.5 (1.6)	10.8 (2.3)	8.5 (2.2)	1.32	0.28
Cognition	21.0 (2.9)	22.1 (3.4)	20.7 (4.1)	18.0 (2.7)	22.8 (7.5)	21.5 (3.5)	20.8 (3.9)	18.8 (2.1)	0.20	0.89
Communication	42.6 (4.9)	34.9 (6.0)	34.1 (4.2)	23.3 (6.3)	35.8 (6.3)	35.0 (4.9)	34.5 (3.7)	30.7 (6.6)	5.63	0.003†
Motivation	24.1 (4.5)	21.6 (4.1)	22.0 (4.7)	18.0 (4.2)	21.2 (5.4)	21.7 (2.9)	21.8 (4.1)	21.0 (2.6)	1.27	0.30
Mannerisms	21.1 (5.2)	16.9 (5.3)	19.9 (4.1)	16.4 (6.2)	22.2 (5.5)	19.0 (3.2)	20.0 (2.1)	18.3 (3.8)	0.27	0.84
ABC score										
Irritability	13.0 (11.6)	10.6 (10.4)	14.4 (14.3)	8.9 (9.9)	8.2 (7.7)	8.3 (6.3)	8.0 (7.1)	9.2 (8.8)	1.78	0.17
Social withdrawal	22.1 (4.6)	13.3 (3.8)	14.4 (8.8)	10.9 (5.1)	18.0 (2.8)	18.2 (7.1)	15.0 (5.1)	17.7 (4.3)	3.08	0.04‡
Stereotypy	5.1 (4.5)	2.4 (2.9)	4.4 (7.5)	3.4 (4.9)	3.8 (4.5)	2.7 (3.8)	4.0 (4.3)	5.2 (5.7)	0.65	0.58
Hyperactivity	15.0 (9.6)	9.0 (8.4)	14.6 (12.6)	12.0 (10.7)	14.2 (14.1)	12.0 (9.4)	15.7 (7.6)	15.3 (8.1)	0.65	0.59
Inappropriate speech	3.7 (3.6)	3.0 (3.1)	4.4 (4.9)	3.7 (3.9)	3.8 (2.1)	3.5 (2.7)	4.0 (2.8)	5.3 (4.1)	0.79	0.51

Treatment group received supplementation with ARA and DHA for 16 weeks.

*Repeated-measures ANOVA was used when baseline scores were used as a covariate, with treatment as the between-subject factor and test session as a repeated factor.

†$P < 0.005$.

‡$P < 0.05$.

TABLE 4. Plasma Levels (Mean±SD) of PUFAs (DHA and ARA), TF, and SOD for the 16-Week Trial

	Groups							
	Supplementation (n = 7)			Placebo (n = 6)				
Measures	Baseline	8 wk	16 wk	Baseline	8 wk	16 wk	F	*P**
PUFAs								
DHA	4.13 (1.50)	4.08 (0.99)	4.11 (1.20)	3.13 (0.97)	3.07 (0.61)	2.96 (0.77)	0.12	0.74
ARA	5.16 (0.77)	6.15 (0.77)	6.36 (0.94)	6.25 (0.95)	5.24 (0.69)	5.34 (0.52	0.03	0.86
Transferrin	228.43 (31.60)	235.00 (40.27)	238.29 (46.15)	261.17 (33.62)	275.50 (35.17)	262.17 (35.64)	7.36	0.03†
SOD	2.73 (0.56)	2.41 (0.51)	2.61 (0.49)	2.32 (0.41)	2.50 (0.51)	2.27 (0.31)	3.77	0.08

*Repeated-measures ANOVA was used when baseline scores were used as a covariate, with treatment as the between-subject factor and test session as a repeated factor.

†$P < 0.05$.

was 1.0. The mechanism of this improvement is not clearly understood. As in previous studies,[15,45] our supplementation regimen did not lead to significant differences in plasma levels of PUFAs. As to the mechanism of action of DHA and ARA, it is well known that ARA preferentially modulates signal transduction,[8,46,47] whereas DHA is an important structural component of neural membranes.[8] Transferrin is involved in signal transduction,[23,24] and SOD plays a role in lipid signaling in defense against oxidative stress.[25,26] Previous reports have suggested that ARA stimulates the production of TF[48] and SOD.[49] Also, a mutual relationship between SOD and TF acts as a mediator in signaling pathways.[50] The present findings regarding the plasma levels of TF, as well as SOD, suggest that signal transduction was up-regulated in the ARA supplementation group.

The present study has several limitations. First, the number of participants was small. Pilot studies generally have small sample sizes, that would provide definitive scientific evidence regarding efficacy. The limitation can be overcome by examining plausible mechanism of action for the treatment/substance undergoing testing.[21] We therefore examined the plasma levels of SOD, TF, and PUFAs. Second, the study included participants with a large age range. The control group included 1 adult who was aged 28 years, but in fact, there was no large discrepancy between the 2 groups. In addition, 12 of the 13 participants had accepted programs that taught appropriate social skills, and 1 male adult worked in a car factory, enabling the development of social peer-based support. Moreover, the 13 participants avoided behavioral intervention and courses that taught appropriate social skills for 3 months before and during the study period. Our results are therefore unlikely to have been affected by the age range or any behavioral interventions. Third, we did not analyze the levels of other antioxidants (eg, tumor necrosis factor α and nitric oxide). Fourth, we found statistically significant changes in only 1 of the 5 subscales of the ABC and SRS. However, the treatment effect study found positive effects of dietary supplementation relative to the placebo group. As the SRS may be more sensitive to changes in measures of social relatedness in ASD,[2] the observed improvement in SRS communication symptoms may be important. Finally, the underlying mechanism of the observed improvement is not clearly understood.

In regard to adverse effects, high doses of arachidonate-enriched triglycerides (SUNTAGA40S) as a source of ARA had no adverse affects on health or growth in rats,[51,52] showing the safety of SUNTAGA40S. Moreover, infant formula containing ARA is reportedly associated with a significantly lower frequency of adverse events than formula without ARA.[53] In this study, no adverse effects were detected in either group, although the small sample size means that the result is not decisive.

In summary, the present preliminary study suggests that supplementation with larger ARA doses added to DHA improves social impairment in individuals with ASD, by up-regulating signal transduction.

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AUTHOR DISCLOSURE INFORMATION

The authors declare no conflicts of interest.

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Erratum: Regarding Article by Nardi et al

In the article by Dr. Antonio E. Nardi, et al titled "A Randomized, Naturalistic, Parallel-Group Study for the Long-Term Treatment of Panic Disorder With Clonazepam or Paroxetine," which was published in the February 2012 issue of the Journal (*J Clin Psychopharmacol* 2012;32:120–126), Figure 1 on page 123 was missing some information. The corrected Figure 1 is reprinted below. We regret this error.

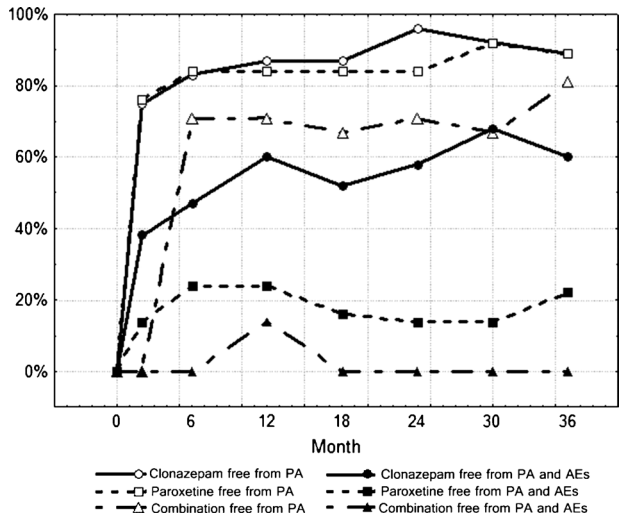

FIGURE 1. Patients free of PAs and AEs throughout long-term treatment.

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Nardo AE, Freire RC, Mochcovitch MD, et al. A randomized, naturalistic, parallel-group study for the long-term treatment of panic disorder with clonazepam or paroxetine. *J Clin Psychopharmacol.* 2012;32:120–126.