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BRIEF COMMUNICATION

Meta-Analysis of BDNF Levels in Autism

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Abstract Brain-derived neurotrophic factor (BDNF) centrally mediates growth, differentiation and survival of neurons, and the synaptic plasticity that underlies learning and memory. Recent meta-analyses have reported significantly lower peripheral BDNF among individuals with schizophrenia, bipolar disorder, and depression, compared with controls. To evaluate the role of BDNF in autism, and to compare autism to psychotic-affective disorders with regard to BDNF, we conducted a meta-analysis of BDNF levels in autism. Inclusion criteria were met by 15 studies, which included 1242 participants. The meta-analysis estimated a significant summary effect size of 0.33 (95 % CI 0.21–0.45, $P < 0.001$), suggesting higher BDNF in autism than in controls. The studies showed notable heterogeneity, but no evidence of publication biases. Higher peripheral BDNF in autism is concordant with several neurological and psychological theories on the causes and symptoms of this condition, and it contrasts notably with the lower levels of BDNF found in schizophrenia, bipolar disorder, and depression.

Keywords Autism · BDNF · Meta-analysis · Schizophrenia · Bipolar disorder · Depression

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Introduction

Brain-derived neurotrophic factor (BDNF) is a protein that belongs to the family of neurotrophins (Brigadski and Lessmann 2014). BDNF regulates dendritic spine maturation and pruning and plays important roles in promoting growth, differentiation, and survival of neurons (Binder and Scharfman 2004; Orefice et al. 2016). Expression of BDNF is regulated in part by neuronal activity induced by sensory stimulation (Woo and Lu 2009), and local protein synthesis at dendrites is mediated by BDNF, whereby it contributes to synaptic plasticity, learning, and memory (Lu et al. 2013; Bowling et al. 2016).

Given the considerable importance of BDNF, levels of this factor have been investigated among individuals with a range of psychiatric conditions. In particular, recent meta-analyses have demonstrated that BDNF levels in serum or plasma are significantly lower in subjects with schizophrenia (Ahmed et al. 2015), bipolar disorder (Fernandes et al. 2015), and depression (Molendijk et al. 2013) than in matched controls. The similar results across these three psychotic-affective disorders are not unexpected, given the strong overlap between them in their causes, phenotypic manifestations, and risk factors (e.g., Konstantareas and Hewitt 2001).

The pattern of association of BDNF levels with autism has been unclear (Tsai 2005; Halepoto et al. 2014). Serum or plasma BDNF is higher among individuals with autism compared with controls in some studies (e.g., Connolly et al. 2006; Ricci et al. 2013), but other studies have reported lower levels (e.g., Nelson et al. 2001; Hashimoto et al. 2007), or nonsignificant differences (e.g., Croen et al. 2008). The overall pattern of association between BDNF and autism has thus remained unresolved, and the causes of

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variation in results among studies have not been investigated.

In this study, we use meta-analytic methods to evaluate the hypothesis of whether peripheral BDNF is altered in autism. We systematically search the literature, conduct a meta-analysis including studies that fit the inclusion criteria, and consider the results in terms of the causes and symptoms of autism, and the relationship of autism with schizophrenia, bipolar disorder, and depression.

Methods

Systematic Literature Search

We searched two databases, PubMed and Web of Science, for peer-reviewed articles up to 11 July 2016. Search terms were "BDNF" AND "Autism," "Plasma" AND "Autism," "Serum" AND "Autism," "Plasma" AND "BDNF," and "Serum" AND "BDNF". Overall, PubMed returned 3506 nonduplicated articles and Web of Science returned 3280.

Studies were included based on the following criteria: (1) they used as subjects individuals with a diagnosis of autism spectrum disorder (ASD); (2) they included measurements of the levels of BDNF in blood; and (3) they provided sufficient data for calculations of effect sizes (group sample size, mean, and standard deviation). Application of the inclusion criteria yielded 17 articles that met all of these stipulations (Consort diagram in Supplementary Fig. 1).

Data Handling

The data in Wang et al. (2015) and Zhang et al. (2014) represent subsets of the data presented in Meng et al. (2016), so only data from the latter study were included. Review of the articles meeting all inclusion criteria showed that seven of them provided insufficient information for computation of effect sizes. We therefore emailed the corresponding authors of these articles. Rodrigues et al. (2014) and Bryn et al. (2015) provided us with the necessary data, but the remaining authors did not reply after multiple attempts. For two of the articles for which authors did not provide data, we extracted data from their figures. For Connolly et al. (2006), the standard deviations were thus measured directly from their Fig. 1, and for Ray et al. (2011), both means and standard deviations were measured from their Fig. 5.

Three studies presented and analyzed multiple groups of autism patients separately (Ray et al. 2011; Ricci et al. 2013; Kasarpalkar et al. 2014), although in each case only one control group was used. For these studies, effect sizes were calculated for each group and averaged, to provide a measure of the study's overall effect size.

Statistical Analysis

Analyses were conducted using R statistical software (version 3.2.4, R Core Team 2015) using the 'metafor' (Viechtbauer 2010) and Hmisc (Harrell 2016) packages. Hedge's g was used as the measure of effect size, adjusted for sample size and within study variance. The fixed effect model was used based on the small number of studies that fit the inclusion criteria and the high methodological homogeneity among the studies. Heterogeneity was measured using the Q test. Publication bias was evaluated using a regression test for funnel plot asymmetry, and using Spearman rank correlation to test for an association between effect sizes and publication years.

Results

Table 1 identifies the studies included in the meta-analysis and includes information about the sample sizes, mean BDNF values, and standard deviation for the autism and control groups. Overall, the analysis included a total of 15 studies and 1242 participants.

The meta-analysis showed that peripheral BDNF levels were moderately increased in subjects with autism when compared with healthy controls ($g = 0.33$, 95 % CI 0.21–0.45, $P < 0.001$). The effect sizes and relative weights of each study, as well as the overall summary effect, are shown in Fig. 1.

The studies demonstrated significant heterogeneity ($Q = 313.02$, $P < 0.01$). One study (Hashimoto et al. 2007) with a large effect size could potentially be considered an outlier. We tested this assumption by removing the study, but no qualitative effect was observed (Q value was not altered substantially, $Q = 252.33$). This heterogeneity may be due in part to differences between studies in the criteria used for autism ascertainment and diagnosis, demographic differences between the populations, and differences in methodology used; measurement of BDNF using serum ($N = 12$ studies) compared with plasma ($N = 3$ studies) did not, however, affect the results (summary effect size 0.35 for serum-based studies, 0.22 for plasma-based studies).

No evidence of publication bias was found using the test for funnel plot asymmetry ($t = -0.114$, $P = 0.91$) (Supplementary Fig. 2). Similarly, no publication bias was observed using Spearman rank correlation between year and study effect size ($r = 0.22$, $P = 0.44$) (Supplementary Fig. 3).

Table 1 Literature and dataset used for meta-analysis and effect sizes

Study		Autism			Control			Effect size
		N	Mean BDNF levels (pg/ml]	SD (Pg/mll	*N*	Mean BDNF levels (pg/ml)	SD (pg/mll	
1	Nelson et al. (2001)	69	13.3	19.9	54	37.4	5	−1.57
2	Miyazaki et al. (2004)	18	25,220	2450	16	17,500	2000	3.35
3	Connolly et al. (2006)	37	32,279	23,379	29	8708	5608	1.30
4	Nelson et al. (2006)	27	3404	1131	20	3299	844	0.10
5	Hashimoto et al. (2007)	18	25,600	2150	18	61,600	10,900	−4.48
e	Croen et al. (2008)	84	54.1	40	159	53.7	47.4	0.01
7	Ray et al. (2011)	21	17.2—mid-to-moderate autism 15.95—severe autism	8.54—mid-to-moderate autism 5.81—severe autism	18	21.72	7.91	−0.78
8	Al-Ayadhi (2012)	44	442	20	40	290	90	2.36
9	Ricci et al. (2013)	29	602.2—mild autism 619.3 moderate autism 791- severe autism	395.1—mild autism 538.0—moderate autism 475.4—severe autism	29	351.4	347.2	0.66
10	Kasarpalkar et al. (2014)	48	198,930—typical ASD 306,680-atypical ASD	65,320—typical ASD 85,930—atypical ASD	29	225,160	79,540	0.29
11	Rodrigues et al. (2014)	28	5198.25	3302.82	19	3883.26	1533.49	0.47
12	Taurines et al. (2014)	24	20.612	6.037	20	24.043	4.34	−0.63
13	Bryn et al. (2015)	65	27,480	12,240	30	21,150	9550	0.55
14	Meng et al. (2016)	82	17,750	5430	82	11,490	2850	1.44
15	Halepoto et al. (2015)	60	392	243	25	290	162	0.45

Unit conversions were performed, as all the means and standard deviations are presented in pg/ml. In the case of Ray et al. (2011), we performed conversions from ng/mg, respectively pg/mg. Due to the fact that the denominators were not expressed in a unit of volume (such as mL), we used a conversion factor for blood density (density = 1.025 g/mL) when performing the unit conversions

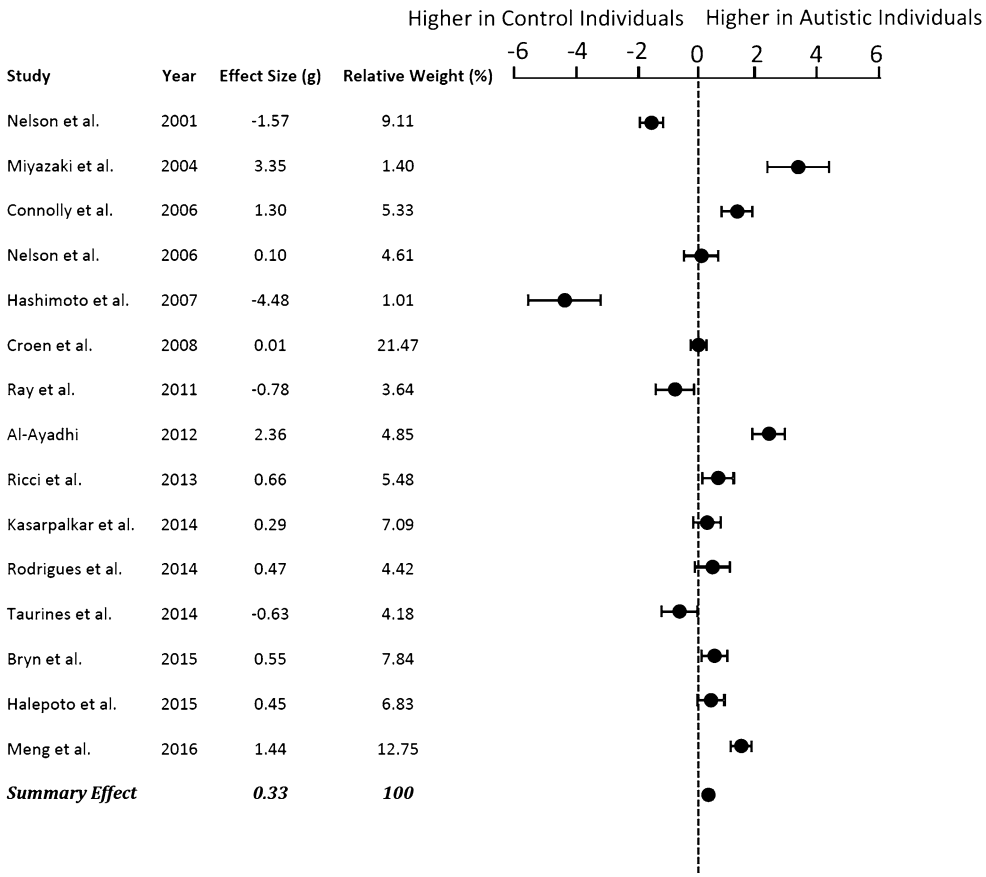

Fig. 1 Effect sizes, CIs, and summary effect size

Discussion

The significant, positive summary effect size of 0.33 reported here, which can be considered of small to medium size, supports the hypothesis that peripheral BDNF levels in autistic subjects are higher than in controls. These results suggest that elevated BDNF may be associated with autism etiology or symptoms, through effects that may involve autism-associated increased protein synthesis at synapses (Kelleher and Bear 2008), enhanced synaptic plasticity (Markram and Markram 2010; Oberman and Pascual-Leone 2014), increased dendritic spine density (Kulkarni and Firestein 2012), or elevated BDNF production mediated by increased sensory sensitivities (Mottron et al. 2006; Baron-Cohen et al. 2009). BDNF levels are also higher than in controls among individuals with Angelman syndrome (Wink et al. 2015), which shows a high incidence of autism (e.g., Bonati et al. 2007), in post-mortem brain tissue (in the fusiform gyrus) of individuals with autism (Garcia et al. 2012), in the prenatal valproic acid and ZnT3-null mouse models of autism (Almeida et al. 2014; Yoo et al. 2016).

The primary limit to interpretation of these meta-analytic results is the heterogeneity found among studies, which cannot be explicated using available information but may involve variation in the autism and control cohorts analyzed, high variability within autism and control groups, and differences in methods used for BDNF quantification (Polacchini et al. 2015). Levels of BDNF have been demonstrated, for example, to vary in some studies in relation to age, sex, or body weight (e.g., Lommatzsch et al. 2005; Iughetti et al. 2011; Pillai et al. 2012; Spratt et al. 2015). Further evaluation of the hypothesis that high BDNF is associated with autism requires robust quantification methods, careful matching of autism and control groups, consideration of cell types and BDNF forms analyzed (Chacón-Fernández et al. 2016), and tests for links of autism diagnoses and autism-related phenotypes with levels of BDNF among clinical and nonclinical individuals.

Our meta-analytic finding of high BDNF in autism stands in notable contrast to the results of recent meta-analyses showing reduced BDNF in schizophrenia (Ahmed et al. 2015), bipolar disorder (Fernandes et al. 2015), and major depression (Molendijk et al. 2013); BDNF is also reduced among individuals with Prader-Willi syndrome, which is caused by opposite genetic and epigenetic alterations to those that cause Angelman syndrome, and which involves high rates of psychosis (Han et al. 2010). Such differences are consistent with a model of autism being diametric (opposite) to psychotic-affective disorders in

major aspects of its causes and phenotypes (Crespi and Badcock 2008; Crespi 2016); for example, schizophrenia is associated with reduced dendritic spine density (Moyer et al. 2015) and deficits in sensory sensitivities (Javitt and Freedman 2015). Evaluation of this hypothesis of opposite alterations would benefit, however, from measurements of BDNF among individuals with autism, and with psychotic-affective disorders, in the same study with identical methods.

References

Ahmed AO, Mantini AM, Fridberg DJ, Buckley PF (2015) Brain-derived neurotrophic factor (BDNF) and neurocognitive deficits in people with schizophrenia: a meta-analysis. Psychiatry Res 226(1):1–13

Al-Ayadhi LY (2012) Relationship between sonic hedgehog protein, brain-derived neurotrophic factor and oxidative stress in autism spectrum disorders. Neurochem Res 37:394–400

Almeida LE, Roby CD, Krueger BK (2014) Increased BDNF expression in fetal brain in the valproic acid model of autism. Mol Cell Neurosci 59:57–62

Baron-Cohen S, Ashwin E, Ashwin C, Tavassoli T, Chakrabarti B (2009) Talent in autism: hyper-systemizing, hyper-attention to detail and sensory hypersensitivity. Phil Trans R Soc London B Biol Sci 364(1522):1377–1383

Binder DK, Scharfman HE (2004) Brain-derived neurotrophic factor. Growth Factors 22(3):123–131

Bonati MT, Russo S, Finelli P, Valsecchi MR, Cogliati F, Cavalleri F, Roberts W, Elia M, Larizza L (2007) Evaluation of autism traits in Angelman syndrome: a resource to unfold autism genes. Neurogenetics 8(3):169–178

Bowling H, Bhattacharya A, Klann E, Chao MV (2016) Deconstructing brain-derived neurotrophic factor actions in adult brain circuits to bridge an existing informational gap in neuro-cell biology. Neural Regen Res 11(3):363–367

Brigadski T, Lessmann V (2014) BDNF: a regulator of learning and memory with clinical relevance. Neuroforum 5:1–11

Bryn V, Halvorsen B, Ueland T, Isaksen J, Kolkova K, Ravn K, Skjeldal OH (2015) Brain derived neurotrophic factor (BDNF) and autism spectrum disorders (ASD) in childhood. Eur J Paediatr Neurol 19(4):411–414

Chacón-Fernández P, Säuberli K, Colzani M, Moreau T, Ghevaert C, Barde Y (2016) Brain-derived neurotrophic factor in megakaryocytes. J Biol Chem 291:9872–9881

Connolly AM, Chez M, Streif EM, Keeling RM, Golumbek PT, Kwon JM, Riviello JJ, Robinson RG, Neuman RJ, Deuel RM (2006) Brain-derived neurotrophic factor and autoantibodies to neural antigens in sera of children with autistic spectrum disorders, Landau-Kleffner syndrome, and epilepsy. Biol Psychiatry 59(4):354–363

Crespi B (2016) The evolutionary etiologies of autism spectrum and psychotic- affective spectrum disorders. Chapter 20. In: Alvergne A (ed) Evolutionary Thinking in Medicine: From Research to Policy and Practice. Oxford University Press, Oxford, pp 299–327

Crespi B, Badcock C (2008) Psychosis and autism as diametrical disorders of the social brain. Behav Brain Sci 31(3):241–261

Croen LA, Goines P, Braunschweig D, Yolken R, Yoshida CK, Grether JK, Fireman B, Kharrazi M, Hansen RL, Van de Water J (2008) Brain-derived neurotrophic factor and autism: maternal and infant peripheral blood levels in the Early Markers for Autism (EMA) study. Autism Res 1(2):130–137

Fernandes BS, Molendjik ML, Kohler CA, Soares JC, Leite CMGS, Machado-Vieira R, Ribeiro TL, Silva JC, Sales PMG, Quevedo J, Oertel-Knochel V, Vieta E, Gonzalez-Pinto A, Berk M, Carvalho AF (2015) Peripheral brain-derived neurotrophic factor (BDNF) as a biomarker in bipolar disorder: a meta-analysis of 52 studies. BMC Med 13:289

Garcia KL, Yu G, Nicolini C, Michalski B, Garzon DJ, Chiu VS, Tongiorgi E, Szatmari P, Fahnestock M (2012) Altered balance of proteolytic isoforms of pro-brain-derived neurotrophic factor in autism. J Neuropathol Exp Neurol 71(4):289–297

Halepoto DM, Bashir S, AL-Ayadhi L (2014) Possible role of brain-derived neurotrophic factor (BDNF) in autism spectrum disorder: current status. J Coll Physicians Surg Pak 24(4):274–278

Halepoto DM, Bashir S, Zeina R, Al-Ayadhi LY (2015) Correlation between Hedgehog (Hh) protein family and brain-derived neurotrophic factor (BDNF) in autism spectrum disorder (ASD). J Coll Physicians Surg Pak 25(12):882–885

Han JC, Muehlbauer MJ, Cui HN, Newgard CB, Haqq AM (2010) Lower brain-derived neurotrophic factor in patients with Prader-Willi syndrome compared to obese and lean control subjects. J Clin Endocrinol Metab 95(7):3532–3536

Harrell FE Jr, with contributions from Dupont C and many others (2016) Hmisc: Harrell Miscellaneous. R package version 3.17-3. https://CRAN.R-project.org/package=Hmisc

Hashimoto K, Iwata Y, Nakamura K, Tsujii M, Tsuchiya KJ, Sekine Y, Suzuki K, Minabe Y, Takei N, Iyo M, Mori N (2007) Reduced serum levels of brain-derived neurotrophic factor in adult male patients with autism. Prog Neuropsychopharmacol Biol Psychiatry 30(8):1529–1531

Javitt DC, Freedman R (2015) Sensory processing dysfunction in the personal experience and neuronal machinery of schizophrenia. Am J Psychiatry 172(1):17–31

Kasarpalkar NJ, Kothari ST, Dave UP (2014) Brain-derived neurotrophic factor in children with Autism Spectrum Disorder. Ann Neurosci 21(4):129–133

Kelleher RJ, Bear MF (2008) The autistic neuron: troubled translation? Cell 135(3):401–406

Konstantareas MM, Hewitt T (2001) Autistic disorder and schizophrenia: diagnostic overlaps. J Autism Dev Discord 31(1):19–28

Kulkarni VA, Firestein BL (2012) The dendritic tree and brain disorders. Mol Cell Neuro 50(1):10–20

Lommatzsch M, Zingler D, Schuhbaeck K, Schloetcke K, Zingler C, Schuff-Werner P, Virchow JC (2005) The impact of age, weight and gender on BDNF levels in human platelets and plasma. Neurobiol Aging 26(1):115–123

Lu B, Nagappan G, Guan X, Nathan PJ, Wren P (2013) BDNF-based synaptic repair as a disease modifying strategy for neurodegenerative diseases. Nat Rev Neurosci 14(6):401–416

Iughetti L, Casarosa E, Predieri B, Patianna V, Luisi S (2011) Plasma brain-derived neurotrophic factor concentrations in children and adolescents. Neuropeptides 45(3):205–211

Markram K, Markram H (2010) The intense world theory—a unifying theory of the neurobiology of autism. Front Hum Neurosci 4:224

Meng WD, Sun SJ, Yang J, Chu RX, Tu W, Liu Q (2016) Elevated serum brain-derived neurotrophic factor (BDNF) but not BDNF gene Val66Met polymorphism is associated with Autism Spectrum Disorders. Mol Neurobiol. doi:10.1007/s12035-016-9721-9

Miyazaki K, Narita N, Sakuta R, Miyahara T, Naruse H, Okado N, Narita M (2004) Serum neurotrophin concentrations in autism and mental retardation: a pilot study. Brain Dev 26(5):292–295

Molendjik ML, Spinhoven P, Polak M, Bus BAA, Penninx BWJH, Elzinga BM (2013) Serum BDNF concentrations as peripheral

manifestations of depression: evidence from a systematic review and meta-analysis on 179 associations (N = 9484). Mol Psychiatry 19(7):791–800

Mottron L, Dawson M, Soulieres I, Hubert B, Burack J (2006) Enhanced perceptual functioning in autism: an update, and eight principles of autistic perception. J Autism Dev Disord 36(1):27–43

Moyer CE, Shelton MA, Sweet RA (2015) Dendritic spine alterations in schizophrenia. Neurosci Lett 5(601):46–53

Nelson KB, Grether JK, Croen LA, Dambrosia JM, Dickens BF, Jelliffe LL, Hansen RL, Phillips TM (2001) Neuropeptides and neurotrophins in neonatal blood of children with autism or mental retardation. Ann Neurol 49(5):597–606

Nelson PG, Kuddo T, Young Song E, Dambrosia JM, Kohler S, Satyanarayana G, VanDunk C, Grether JK, Nelson KB (2006) Selected neurotrophins, neuropeptides, and cytokines: developmental trajectory and concentrations in neonatal blood of children with autism or Down syndrome. Int J Dev Neurosci 24:73–80

Oberman LM, Pascual-Leone A (2014) Hyperplasticity in autism spectrum disorder confers protection from Alzheimer's disease. Med Hypotheses 83:337–342

Orefice LL, Shih CC, Xu H, Waterhouse EG, Xu B (2016) Control of spine maturation and pruning through proBDNF synthesized and released in dendrites. Mol Cell Neurosci 71:66–79

Pillai A, Bruno D, Sarreal AS, Hernando RT, Saint-Louis LA, Nierenberg J, Ginsberg SD, Pomara N, Mehta PD, Zetterberg H, Blennow K, Buckley PF (2012) Plasma BDNF levels vary in relation to body weight in females. PLoS One 7(7):e39358

Polacchini A, Metelli G, Francavilla R, Baj G, Florean M, Mascaretti LG, Tongiorgi E (2015) A method for reproducible measurements of serum BDNF: comparison of the performance of six commercial assays. Sci Rep 5:17989

Ray B, Long JM, Sokol DK, Lahiri DK (2011) Increased secreted amyloid precursor protein-a (sAPPa) in severe autism: proposal of a specific, anabolic pathway and putative biomarker. PLoS ONE 6(6):e20405

Ricci S, Businaro R, Ippoliti F, Lo Vasco VR, Massoni F, Onofri E, Troili GM, Pontecorvi V, Morelli M, Rapp Ricciardi M, Archer T (2013) Altered cytokine and BDNF levels in autism spectrum disorder. Neurotox Res 24(4):491–501

Rodrigues DH, Rocha NP, Sousa LF, Barbosa IG, Kummer A, Teixeira AL (2014) Circulating levels of neurotrophic factors in autism spectrum disorders. Neuro Endocrinol Lett 35(5):380–384

Spratt EG, Granholm A-C, Carpenter LA, Boger HA, Papa CE, Logan S, Chaudhary H, Boatwright S-W, Brady KT (2015) Pilot study and review: physiological differences in BDNF, a potential biomarker in males and females with autistic disorder. Int Neuropsychiatr Dis J 3:19–26

Taurines R, Segura M, Schecklmann M, Albantakis L, Grünblatt E, Walitza S, Jans T, Lyttwin B, Haberhausen M, Theisen FM, Martin B, Briegel W, Thome J, Schwenck C, Romanos M, Gerlach M (2014) Altered peripheral BDNF mRNA expression and BDNF protein concentrations in blood of children and adolescents with autism spectrum disorder. J Neural Transm 121(9):1117–1128

R Core Team (2015) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna. http://www.R-project.org/

Tsai SJ (2005) Is autism caused by early hyperactivity of brain-derived neurotrophic factor? Med Hypotheses 65(1):79–82

Viechtbauer W (2010) Conducting meta-analyses in R with the metafor package. J Stat Software, 36(3):1–48. http://www.jstatsoft.org/v36/i03/

Wang M, Chen H, Yu T, Cui G, Jiao A, Liang H (2015) Increased serum levels of brain-derived neurotrophic factor in autism spectrum disorder. Neuro Rep 26(11):638–641

Wink LK, Fitzpatrick S, Shaffer R, Melnyk S, Begtrup AH, Fox E, Schaefer TL, Mathieu-Frasier L, Ray B, Lahiri D, Horn PA (2015) The neurobehavioral and molecular phenotype of Angelman Syndrome. Am J Med Genet Part A 167(11):2623–2628

Woo NH, Lu B (2009) BDNF in synaptic plasticity and memory. In: Malenka R (ed) Intracellular communication In the nervous system. Bethesda, Maryland, pp 135–143

Yoo MH, Kim T-Y, Yoon YH, Koh J-Y (2016) Autism phenotypes in ZnT3 null mice: involvement of zinc dyshomeostasis, MMP-9 activation and BDNF upregulation. Sci Rep 6:28548

Zhang QB, Jiang LF, Lu YJ (2014) Serum brain-derived neurotrophic factor levels in Chinese children with autism spectrum disorders: a pilot study. Int J Dev Neurosci 37:65–68